

SAFRAMYCINS, ANALOGUES AND USES THEREOF

PRIORITY INFORMATION

The present application claims priority under 35 U.S.C. § 119(e) to provisional
5 application number 60/245,888, filed November 3, 2000, entitled "Synthesis of Saframycins,
Analogues and Uses Thereof", the entire contents of which are hereby incorporated by reference.

GOVERNMENT SUPPORT

This invention was made in part with a grant from the National Institutes of Health
10 (Grant Number: 7 R37 CA47148-12). Therefore, the government has certain rights in the
invention.

BACKGROUND OF THE INVENTION

The discovery of novel therapeutic agents has traditionally relied on the identification of
15 biologically active secondary metabolites of microorganisms. These compounds have provided a
rich source of natural products that have either been utilized directly as effective therapeutic
agents, or have provided leads for novel therapeutic agents to be developed through synthetic
techniques.

One disease for which the development of novel therapeutics is particularly important is
20 cancer, which not only has eluded a "cure", but is also one of the leading disease-related causes
of death of the human population. Examples of anticancer agents that have been identified from
or developed from natural sources include paclitaxel, mitomycin C, and adriamycin to name a
few. One drawback to the use of secondary metabolites from natural resources, however, has
been that these agents are generally only present in minute quantities. Fortunately, in an effort to
25 make these agents more available for use, and to enable further pharmaceutical research,
synthetic chemists have developed elegant and efficient synthetic strategies to enable the
production of either the natural products themselves, or useful derivatives thereof.

Although these therapeutic agents, and others developed from natural sources, through
the efforts of synthetic chemistry, are currently in use for the treatment of individuals having
30 cancer, many of these agents, as well as other common treatments such as surgery and radiation,
are often unselective for tumor cells and/or are so toxic as to render the individual significantly

immunocompromised. Thus, although many strides have been made in the development of novel treatments, there remains a need for the identification of additional therapeutics, preferably those that are more selective and less toxic.

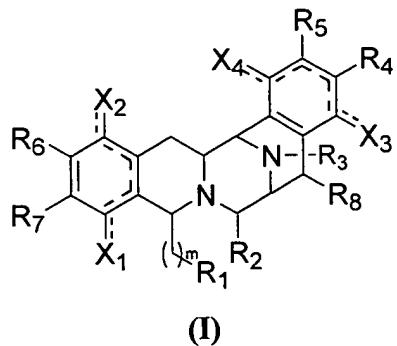
One particular family of natural products that has generated significant interest is the saframycins. The saframycins are a class of antibiotics with activity against gram-positive bacteria and also against several kinds of tumor. Specifically, several saframycin analogues have been isolated and characterized in recent years (see, DE 2839668; US Patents 4,248,863; 4,372,947; 5,023,184; 4,837,149; and EP 329606). For example saframycins A-H, R and S have been isolated from the culture broths of *Streptomyces lavendulae*, and saframycins M_{x1} and M_{x2}, have been isolated from the culture broths of the myxobacterium, *Myxococcus xanthus*, each of the saframycins varying in the oxidation state of the ring system and in substitution of the core structure (see, for example, Saito *et al. Chem. Pharm. Bull.* **1995**, *43*, 777). It has been suggested that certain saframycins, namely A and C exhibit extreme cytotoxicity toward culture cells and toward several experimental tumors including leukemias L1210 and P388 and Ehrlich carcinoma. Specifically, saframycin A has been shown to block RNA synthesis in cultured cells, and it has been suggested that saframycins A and C exhibit this potency because of their ability to bind and cleave DNA (for a discussion of the biological activity of saframycins see, for example, Lown *et al. Biochemistry* **1982**, *21*, 419; Ishiguro *et al. Biochemistry* **1978**, *17*, 2545; Rao *et al. Chem. Res. Toxicol.* **1990**, *3*, 262; Ishiguro *et al. J Biol. Chem.* **1981**, *256*, 2162).

Although this class of natural products has shown promising biological activity, there have been few investigations into the synthesis and development of novel analogues of this family of natural products (see, EP 233841; EP 173649; Fukuyama *et al. J. Am. Chem. Soc.* **1982**, *104*, 4957-4958; Kubo *et al. J. Org. Chem.* **1988**, *53*, 4295-4310; Fukuyama *et al. J. Am. Chem. Soc.* **1990**, *112*, 3712-3713).

Clearly, there remains a need to further investigate the potential of this class of natural products, and analogues thereof, to develop novel, more potent and more selective therapeutics. Additionally, because of the complexity of the structure of this class of natural products, there also remains a need to develop additional synthetic techniques to rapidly access novel compounds based upon the general core structure of the saframycins, and other related compounds.

SUMMARY OF THE INVENTION

In recognition of the need to develop novel therapeutic agents and efficient methods for the synthesis thereof, the present invention provides novel compounds of general formula (I), and methods for the synthesis thereof.



The present invention additionally provides pharmaceutical compositions comprising a compound of formula (I) and a pharmaceutically acceptable carrier. In yet another aspect, the
10 present invention provides methods for treating cancer comprising administering a therapeutically effective amount of a compound of formula (I) to a subject in need thereof.

DESCRIPTION OF THE FIGURES

Figure 1 depicts a retrosynthetic analysis of saframycin A.

15 Figure 2 depicts the synthesis of saframycin A and precursors. Reaction conditions: (a) Na₂SO₄, CH₂Cl₂, 23°C, >90%; LiBr, DME, 35 C, 65-72%. (b) CH₂O-H₂O, NaBH(OAc)₃, CH₃CN, 23°C, 94%. (c) HOAc, TBAF, THF, 23°C; DBU, CH₂Cl₂, 23°C, 92%. (d) Na₂SO₄, CH₂Cl₂, 23°C, 66%. (e) ZnCl₂, TMSCN, CF₃CH₂OH-THF, 23°C, 86%. (f) DBU, CH₂Cl₂, 23°C, 88%. (g) ClCOCOCH₃, PhNEt₂, CH₂Cl₂, 0°C, 89%. (h) PhIO, CH₃CN-H₂O, 0°C, 66%.

20 Figure 3 depicts the synthesis of a precursor of saframycin A (9) from an N-linked trimeric α-amino aldehyde precursor.

Figure 4 depicts precursors in the synthesis of saframycin A.

Figure 5 depicts the synthesis of an N-linked trimeric α-amino aldehyde precursor (8).

25 Figure 6 depicts the synthesis of a precursor of saframycin A (9) from an N-linked trimeric α-amino aldehyde precursor (8).

Figure 7 depicts the synthesis of exemplary inventive analogues of saframycin.

Figure 8 depicts the synthesis of exemplary inventive analogues of saframycin.

Figure 9 depicts the synthesis of exemplary inventive analogues of saframycin.

Figure 10 depicts a general method for the rapid synthesis of large numbers of potent derivatives of saframycin A.

5 Figure 11 depicts the solid phase synthesis of derivatives of saframycin A. Reaction conditions: (a) Imidazole, DMF, 23°C; CH₃OH, Imidazole, 23°C, 100% (2 steps). (b) TBAF, AcOH, THF, 23°C. (c) Piperidine, DMF, 23°C. (d) aldehyde 1, DMF, 23°C. (e) LiBr, DME, 35°C, 83% (4 steps). (f) CH₂O-H₂O, NaBH(OAc)₃, DMF, 23°C, 95%. (g) Fmoc-glycinal, DCE, 40°C, 80% (3 steps). (h) ZnCl₂, TMSCN, CF₃CH₂OH-THF, 23°C, 48%. (* = 4.5:1 mixture of 10 diastereomers (cis- is major))

10 Figure 12 depicts the synthesis of the siloxymorpholine reagent (5). Reaction conditions: (a) Jacobsen HKR (Tokunaga, M.; Larrow, J. F.; Kakiuchi, F.; Jacobsen, E. N. *Science*. 1997, 277, 936–938). 23 °C, 100% (98% ee). (b) 2-aminoethanol, EtOH, 0→70 °C, 100%. (c) BnBr, KHCO₃, DMF, 50 °C, 97%. (d) NaH, THF, 0→23 °C; TsIm, THF, 0→23 °C, 70% (2 steps, 15 >95% ee). (e) 10% Pd/C, H₂, CH₃OH, AcOH, 23 °C, 96%.

15 Figure 13 depicts reaction with a substituted aldehyde reagent (via Pictet-Spengler reaction) to generate diversified pentacyclic core structures.

20 Figures 14A and 14B depict a synthetic scheme illustrating the generation of diversity in the R₃ position of generic structure (I) (via alternate N-alkylation reaction).

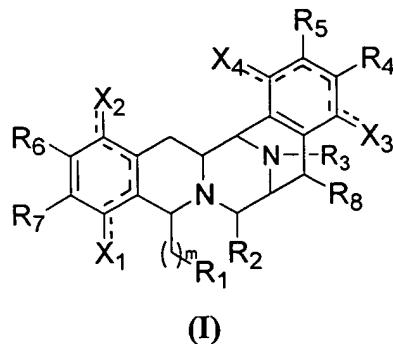
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DETAILED DESCRIPTION OF THE INVENTION

In recognition of the need to develop novel anticancer therapeutics and more efficient processes for the preparation of such therapeutics, the present invention provides novel compounds and methods for the preparation thereof. In general, in one aspect, the present 25 invention provides novel analogues of the saframycin antitumor antibiotics having anticancer activity. In yet another aspect, the present invention provides efficient methods for the generation of these compounds, and alkaloids in general, involving the directed condensation of substituted aldehyde precursors. Significantly, the methodology provided by the present invention enables the efficient production of these novel compounds in significant quantities for 30 therapeutic use.

1) General Description of Compounds of the Invention

As mentioned above, in one aspect of the invention, novel analogues of the saframycin antitumor antibiotics are provided. In general, compounds having the structure (I) are provided:



wherein R₁ is NR_AR_B, -OR_A, -SR_A, -C(=O)R_A, -C(=S)R_A, -S(O)₂R_A, or an aliphatic, heteroaliphatic, aryl, heteroaryl, (aliphatic)aryl, (aliphatic)heteroaryl, (heteroaliphatic)aryl, or (heteroaliphatic)heteroaryl moiety, wherein each occurrence of R_A and R_B is independently hydrogen, -(C=O)R_C, -NHR_C, -(SO₂)R_C, -OR_C, or an aliphatic, heteroaliphatic, aryl, or heteroaryl moiety, or R_A and R_B, when taken together form an aryl, heteroaryl, cycloaliphatic, or cycloheteroaliphatic moiety, wherein each occurrence of R_C is independently hydrogen, -ORD, -SR_D, -NHR_D, -(C=O)R_D, or an aliphatic, heteroaliphatic, aryl, or heteroaryl moiety, wherein each occurrence of R_D is independently hydrogen, a protecting group, or an aliphatic, heteroaliphatic, aryl, heteroaryl, acyl, alkoxy, aryloxy, alkylthio, arylthio, heteroaryloxy, or heteroarylthio moiety;

wherein R₂ is hydrogen, -OR_E, =O, -C(=O)R_E, -CO₂R_E, -CN, -SCN, halogen, -SR_E, -SOR_E, -SO₂R_E, -NO₂, -N(R_E)₂, -NHC(O)R_E, or an aliphatic, heteroaliphatic, aryl, or heteroaryl moiety, wherein each occurrence of R_E is independently hydrogen, a protecting group, or an aliphatic, heteroaliphatic, aryl, heteroaryl, acyl, alkoxy, aryloxy, alkylthio, arylthio, heteroaryloxy, or heteroarylthio moiety;

wherein R₃ is hydrogen, a nitrogen protecting group, -COOR_F, -COR_F, -CN, or an aliphatic, heteroaliphatic, aryl, or heteroaryl moiety, wherein each occurrence of R_F is independently hydrogen, a protecting group, or an aliphatic, heteroaliphatic, aryl, heteroaryl, alkoxy, aryloxy, alkylthio, arylthio, heteroaryloxy, or heteroarylthio moiety;

wherein R₄ and R₆ are each independently hydrogen, or an aliphatic, heteroaliphatic, aryl, heteroaryl, acyl, alkoxy, aryloxy, alkylthio, arylthio, heteroaryloxy, or heteroarylthio moiety;

wherein R₅ and R₇ are each independently hydrogen, -OR_G, -C(=O)R_G, -CO₂R_G, -CN, -SCN, halogen, -SR_G, -SOR_G, -SO₂R_G, -NO₂, -N(R_G)₂, -NHC(O)R_G, or an aliphatic, heteroaliphatic, aryl or heteroaryl moiety, wherein each occurrence of R_G is independently hydrogen, a protecting group, or an aliphatic, heteroaliphatic, aryl, heteroaryl, acyl, alkoxy, aryloxy, alkylthio, arylthio, heteroaryloxy, or heteroarylthio moiety;

wherein R₈ is hydrogen, alkyl, -OH, protected hydroxyl, =O, -CN, -SCN, halogen, -SH, protected thio, alkoxy, thioalkyl, amino, protected amino, or alkylamino;

10 wherein m is 0-5;

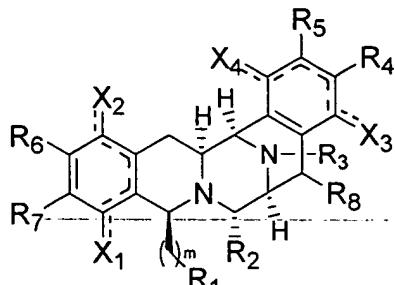
wherein X₁, X₂, X₃ and X₄ are each independently hydrogen, -OR_H, =O, -C(=O)R_H, -CO₂R_H, -CN, -SCN, halogen, -SR_H, -SOR_H, -SO₂R_H, -NO₂, -N(R_H)₂, -NHC(O)R_H, or an aliphatic, heteroaliphatic, aryl, or heteroaryl moiety, wherein each occurrence of R_H is independently hydrogen, a protecting group, or an aliphatic, heteroaliphatic, aryl, heteroaryl, acyl, alkoxy, aryloxy, alkylthio, arylthio, heteroaryloxy, or heteroarylthio moiety;

15 whereby if at least either X₁ and X₂ or X₃ and X₄ are doubly bonded to the 6-membered ring, then the dotted bonds in either or both of the 6-membered rings represent two single bonds and one double bond, and a quinone moiety is generated, or if at least either X₁ and X₂ or X₃ and X₄ are singly bonded to the 6-membered ring, then the dotted bonds in either or both of the 6-membered rings represent two double bonds and one single bond, and a hydroquinone moiety is generated;

20 whereby each of the foregoing aliphatic, heteroaliphatic and alkyl moieties may independently be substituted or unsubstituted, branched or unbranched, or cyclic or acyclic, and each of the foregoing aryl or heteroaryl moieties may independently be substituted or unsubstituted; and

25 pharmaceutically acceptable derivatives thereof.

In certain embodiments of the invention, compounds of formula (I) have the following stereochemistry and structure as shown in formula (Ia):



(Ia)

It will be appreciated that, in certain embodiments of the compounds as described
5 generally above and in classes and subclasses herein certain naturally occurring saframycins and other related natural products are excluded including:

saframycins A, B, C, D, E, F, G, H, R, S₁, Y₃, Y_{d1}, A_{d1}, Y_{d2}, Y_{2b}, Y_{2b-d}, AH₂, AH₂Ac, AH₁, AH₁Ac, AR₃, M_{x-1} and M_{x-2}; safracins A and B; reineramycins A, B, D, E, and F; and xestomycin.

10

In certain other embodiments, for compounds as described above and in subclasses herein, when m is 1, R₁ excludes any one or more of the following groups: -NH(protecting group), -NH₂, -NHCOCOMe, -NHCOC(Me)(OMe)(OMe), -NHCOCH(NH₂)CH₃, -NHCOCH(NH(acyl))CH₃ -NHCOCH(NH₂)Ac, or NHCOCH(NHCOOBn)(Me);
15 O(C=O)C(CH₃)=C(CH₃)H; -OH, -O(protecting group), -O(COCH₃), -O(C=O)CH₂CH₃.

In still other embodiments, for certain of the compounds as described above and herein, when m is 1; when X₁, X₂, X₃ and X₄ are each =O; when R₂ is -CN or -OH; when R₄ and R₆ are each -CH₃; when R₅ and R₇ are each -OCH₃; when R₈ is H; and R₁ is -NH(C=O)R_C, then R_C is not -CH(NR_WR_Y)(CH₂R_Z) where R_W and R_Y are each independently hydrogen or C₁₋₇ alkyl, 20 aryl(C₁₋₄)alkyl, (C₁₋₄)alkylaryl, a substituted sulfonyl (-S(O)₂-) group, or a substituted acyl group, and where R_Z is hydrogen or C₁₋₄ alkyl.

In yet other embodiments, for certain of the compounds as described above and herein, 25 when m is 1; when X₁, X₂, X₃ and X₄ are each =O; when R₂ is -CN; when R₄ and R₆ are each -

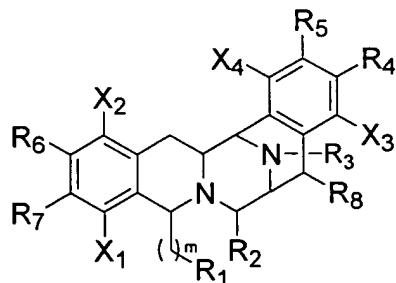
CH₃; when R₅ and R₇ are each -OCH₃; when R₈ is H; and R₁ is -NH(C=O)R_C, then R_C is not -C(OH)(Me)CH₂(C=O)Me.

In still other embodiments, for certain of the compounds as described above and herein,
5 when m is 1 and when R₂ is H; and R₁ is -NH(C=O)R_C, then R_C is not -CH(Me)NH(C=O)O(CH₂)Ph.

In yet other embodiments, for certain of the compounds as described above and herein,
when m is 0; R₂ is H; X₃ is H; and R₁ is -C(=O)R_A, then R_A is not -O(alkyl). Alternatively, in
10 certain other embodiments, when R₂ is H; m is 1; and R₁ is -OR_A, then R_A is not -C(=O)R_C, or
S(O)₂R_C, wherein R_C is an alkyl moiety.

2) Featured Classes of Compounds

It will be appreciated that for compounds as generally described above, certain classes of
15 compounds are of special interest. For example, one class of compounds of special interest
includes those compounds of the invention as described above and in certain subclasses herein,
wherein the compounds have the general structure (II):

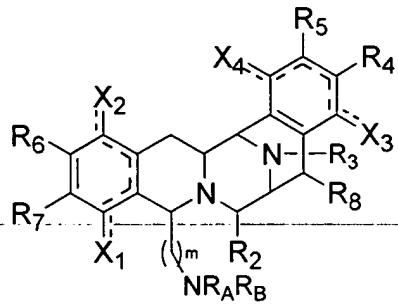


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(II)

wherein R₁-R₈, X₁-X₄ and m are as defined above and in subclasses herein.

25 Another class of compounds of special interest includes those compounds of the
invention as described above and in certain subclasses herein, wherein the compounds have the
general structure (III):

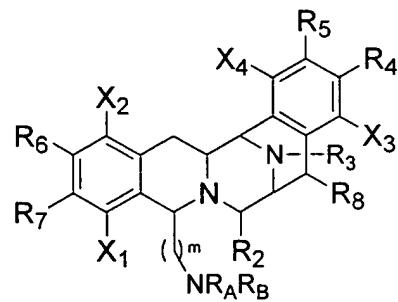


(III)

wherein R₂-R₈, X₁-X₄, m, R_A and R_B are as defined above and in subclasses herein.

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Yet another class of compounds of special interest includes those compounds of the invention as described above and in certain subclasses herein, wherein the compounds have the general structure (IV):

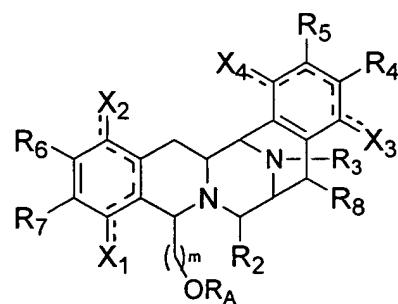


(IV)

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wherein R₂-R₈, X₁-X₄, m, R_A and R_B are as defined above and in subclasses herein.

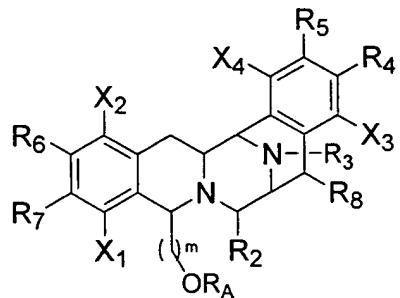
Yet another class of compounds of special interest includes those compounds of the
15 invention as described above and in certain subclasses herein, wherein the compounds have the general structure (V):



(V)

wherein R₂-R₈, X₁-X₄, m, and R_A are as defined above and in subclasses herein.

5 Yet another class of compounds of special interest includes those compounds of the invention as described above and in certain subclasses herein, wherein the compounds have the general structure (VI):

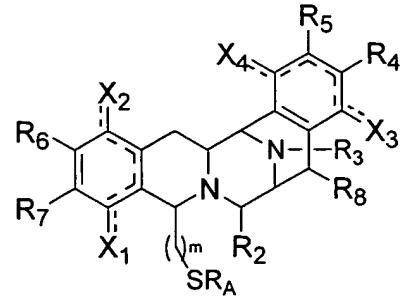


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(VI)

wherein R₂-R₈, X₁-X₄, m, and R_A are as defined above and in subclasses herein.

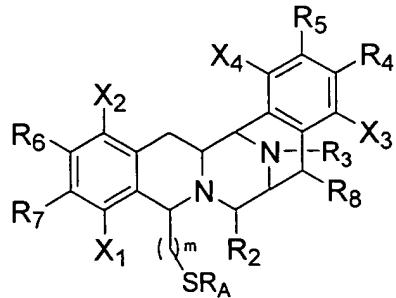
15 Yet another class of compounds of special interest includes those compounds of the invention as described above and in certain subclasses herein, wherein the compounds have the general structure (VII):



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wherein R₂-R₈, X₁-X₄, m, and R_A are as defined above and in subclasses herein.

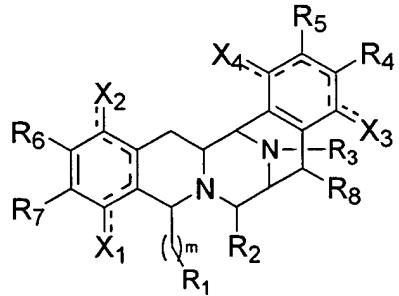
Yet another class of compounds of special interest includes those compounds of the invention as described above and in certain subclasses herein, wherein the compounds have the general structure (VIII):



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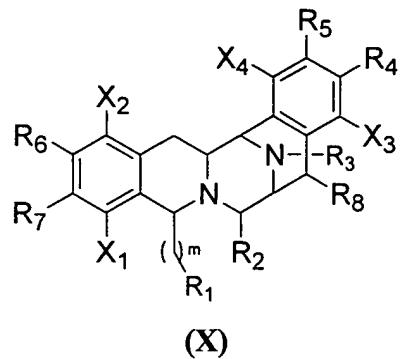
wherein R₂-R₈, X₁-X₄, m, and R_A are as defined above and in subclasses herein.

Still another class of compounds of special interest includes those compounds of the
10 invention as described above and in certain subclasses herein, wherein the compounds have the
general structure (IX):



15 wherein R₂-R₈, X₁-X₄, and m are as defined above and in subclasses herein, and wherein
R₁ is a substituted or unsubstituted, cyclic or acyclic, branched or unbranched aliphatic or
heteroaliphatic moiety, or is a substituted or unsubstituted aryl or heteroaryl moiety.

20 Yet another class of compounds of special interest includes those compounds of the
invention as described above and in certain subclasses herein, wherein the compounds have the
general structure (X):

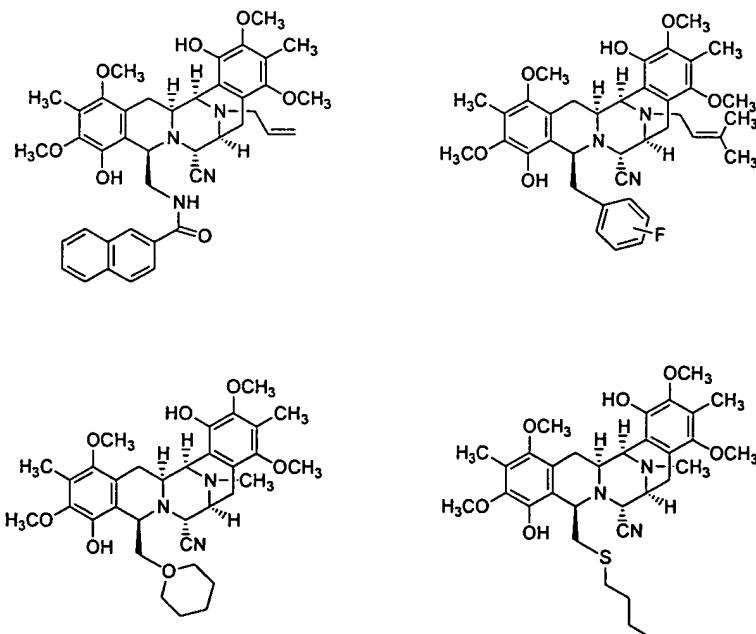


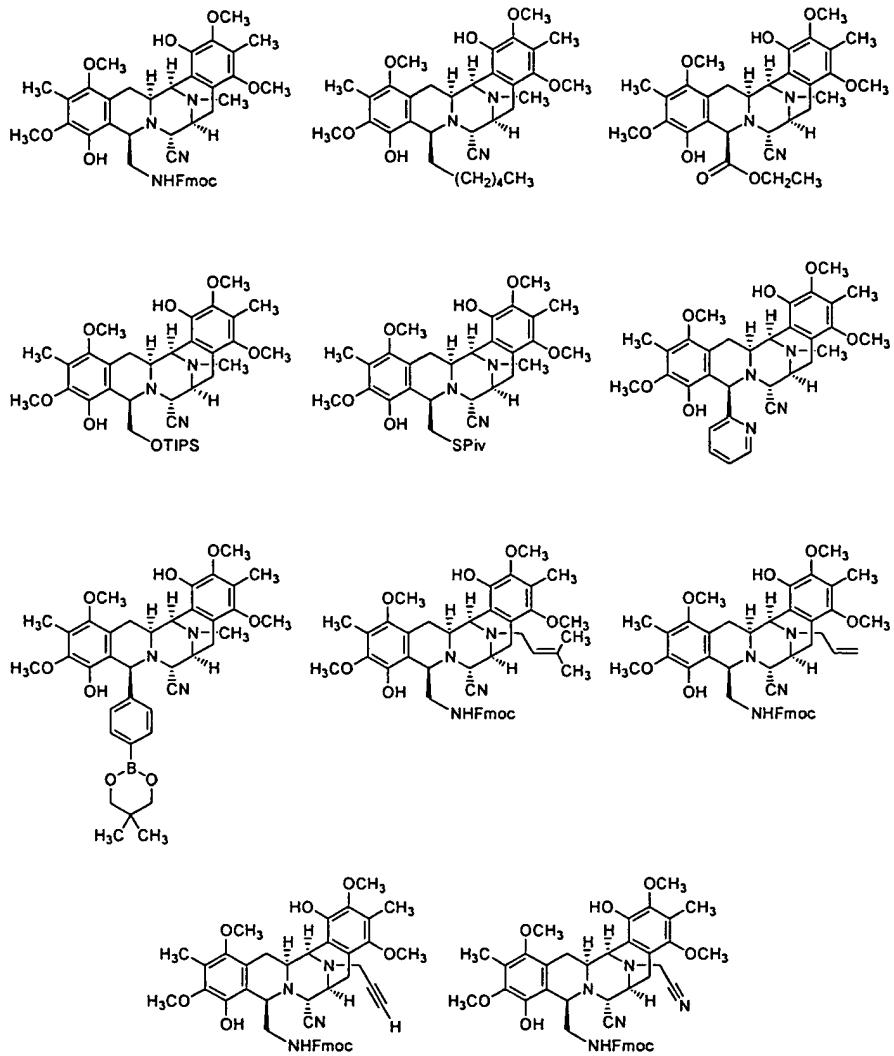
wherein R₂-R₈, X₁-X₄, and m are as defined above and in subclasses herein, and wherein

5 R₁ is a substituted or unsubstituted, cyclic or acyclic, branched or unbranched aliphatic or heteroaliphatic moiety, or is a substituted or unsubstituted aryl or heteroaryl moiety.

The following compounds are illustrative of certain of the compounds described generally and in classes and subclasses herein:

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A number of important subclasses of each of the foregoing classes deserve separate
5 mention; these subclasses include subclasses of each of the foregoing classes in which:

- i) compounds of the invention as described above and herein are enantiopure;
- ii) compounds as described above and in subclasses herein, wherein when m is 1, R₁ excludes any one or more of the following groups: -NH(protecting group), -NH₂, -NHCOCOMe,
10 -NHCOC(Me)(OMe)(OMe), -NHCOCH(NH₂)CH₃, -NHCOCH(NH(acyl))CH₃ -
NHCOCH(NH₂)Ac, or NHCOCH(NHCOOBn)(Me); -O(C=O)C(CH₃)=C(CH₃)H; -OH, -
O(protecting group), -O(COCH₃), -O(C=O)CH₂CH₃;

iii) compounds as described above and in subclasses herein, wherein when m is 1; when X₁, X₂, X₃ and X₄ are each =O; when R₂ is -CN or -OH; when R₄ and R₆ are each -CH₃; when R₅ and R₇ are each -OCH₃; when R₈ is H; and R₁ is -NH(C=O)R_C, then R_C is not -CH(NR_WR_Y)(CH₂R_Z) where R_W and R_Y are each independently hydrogen or C₁₋₇ alkyl, aryl(C₁₋₄)alkyl, (C₁₋₄)alkylaryl, a substituted sulfonyl (-S(O)₂-) group, or a substituted acyl group, and where R_Z is hydrogen or C₁₋₄ alkyl;

iv) compounds as described above and in subclasses herein, wherein when m is 1; when X₁, X₂, X₃ and X₄ are each =O; when R₂ is -CN; when R₄ and R₆ are each -CH₃; when R₅ and R₇ are each -OCH₃; when R₈ is H; and R₁ is -NH(C=O)R_C, then R_C is not -

5 10 C(OH)(Me)CH₂(C=O)Me;

v) compounds as described above and in subclasses herein, wherein when m is 1 and when R₂ is H; and R₁ is -NH(C=O)R_C, then R_C is not -CH(Me)NH(C=O)O(CH₂)Ph;

vi) compounds as described above and in subclasses herein, wherein when m is 0; R₂ is H; X₃ is H; and R₁ is -C(=O)R_A, then R_A is not -O(alkyl). Alternatively, in certain other 15 embodiments, when R₂ is H; m is 1; and R₁ is -OR_A, then R_A is not -C(=O)R_C, or S(O)₂R_C, wherein R_C is an alkyl moiety.

vii) m is 0 or 1;

viii) R₂ is CN, -SCN, =O, OH, protected hydroxyl, H, or alkoxy;

ix) R₂ is hydrogen, hydroxyl, -CN or -SCN;

20 x) R₃ is hydrogen, a nitrogen protecting group, -CN, -CH₂CN, aliphatic or aryl;

xi) R₄ and R₆ are each alkyl;

xii) R₅ and R₇ are each alkyloxy or thioalkyl;

xiii) R₈ is hydrogen, alkyl, -OH, protected hydroxyl, =O, -CN, -SCN, halogen, -SH, protected thio, alkoxy, thioalkyl, amino, protected amino, or alkylamino;

25 xiv) R₈ is hydrogen;

xv) X₁, X₂, X₃, and X₄ are each independently alkoxy, -OH, protected hydroxyl, or =O;

xvi) R₂ is CN, -SCN, =O, OH, protected hydroxyl, H, or alkoxy; R₃ is hydrogen, a nitrogen protecting group, -CN, -CH₂CN, aliphatic, or aryl; R₄ and R₆ are each alkyl; R₅ and R₇ are each alkyloxy or thioalkyl; R₈ is hydrogen, alkyl, -OH, protected hydroxyl, =O, CN, halogen, SH, alkoxy, thioalkyl, amino, or alkylamino; and X₁, X₂, X₃, and X₄ are each independently alkoxy, OH or =O;

xvii) R_2 is -CN, -SCN, -OH, protected hydroxyl, H, or alkoxy; R_3 is hydrogen, a nitrogen protecting group, aliphatic, or aryl; R_4 and R_6 are each alkyl; R_5 and R_7 are each alkyloxy or thioalkyl; X_1 and X_4 are each -OH; R_8 is hydrogen, alkyl, OH, protected hydroxyl, =O, CN, halogen, SH, alkoxy, thioalkyl, amino, or alkylamino; and X_2 and X_3 are each alkyloxy or

5 thioalkyl;

xviii) X_1 is OH, X_2 is OCH₃, X_3 is OCH₃, X_4 is OH, R_2 is CN, H or OH, R_3 is Me, R_4 is CH₃, R_5 is OCH₃, R_6 is CH₃, R_7 is OCH₃, and R_8 is H;

xix) R_1 is OR_A, SR_A, or NR_AR_B, wherein R_A and R_B are each independently hydrogen, -(C=O)R_C or an aliphatic, heteroaliphatic, aryl, or heteroaryl moiety, wherein R_C is -(C=O)R_D, or 10 an aliphatic, heteroaliphatic, aryl or heteroaryl moiety, and wherein R_D is an aliphatic, heteroaliphatic, aryl, or heteroaryl moiety, or wherein R_A and R_B, taken together, form a heterocyclic moiety,

whereby each of said aliphatic and heteroaliphatic moieties is independently substituted or unsubstituted, branched or unbranched, or cyclic or acyclic, and each of said aryl, 15 heteroaryl and heterocyclic moieties is independently substituted or unsubstituted;

xx) R_1 is OR_A, SR_A, or NR_AR_B, wherein R_A and R_B are each independently hydrogen, -(C=O)R_C, or an aryl, (aliphatic)aryl, (heteroaliphatic)aryl, heteroaryl, (aliphatic)heteroaryl, or (heteroaliphatic)heteroaryl moiety, wherein R_C is an aryl, (aliphatic)aryl, (heteroaliphatic)aryl, 20 heteroaryl, (aliphatic)heteroaryl, or (heteroaliphatic)heteroaryl moiety, or wherein R_A and R_B taken together form a heterocyclic moiety,

whereby each of said aliphatic and heteroaliphatic moieties is independently substituted or unsubstituted, branched or unbranched, or cyclic or acyclic, and each of said aryl, heteroaryl and heterocyclic moieties is independently substituted or unsubstituted;

xxi) R_1 is -NR_AC(=O)R_C, wherein R_A is hydrogen or lower alkyl, and R_C is a substituted 25 or unsubstituted, branched or unbranched, cyclic or acyclic aliphatic or heteroaliphatic moiety, or a substituted or unsubstituted aryl or heteroaryl moiety, or wherein R_A and R_C taken together form a heterocyclic or heteroaryl moiety;

xxii) R_1 is NR_AC(=O)R_C, wherein R_A is hydrogen or lower alkyl, and R_C is an aryl, (aliphatic)aryl, (aliphatic)heteroaryl, heteroaryl, (heteroaliphatic)aryl, or 30 (heteroaliphatic)heteroaryl moiety, or wherein R_A and R_C taken together form a heterocyclic or heteroaryl moiety;

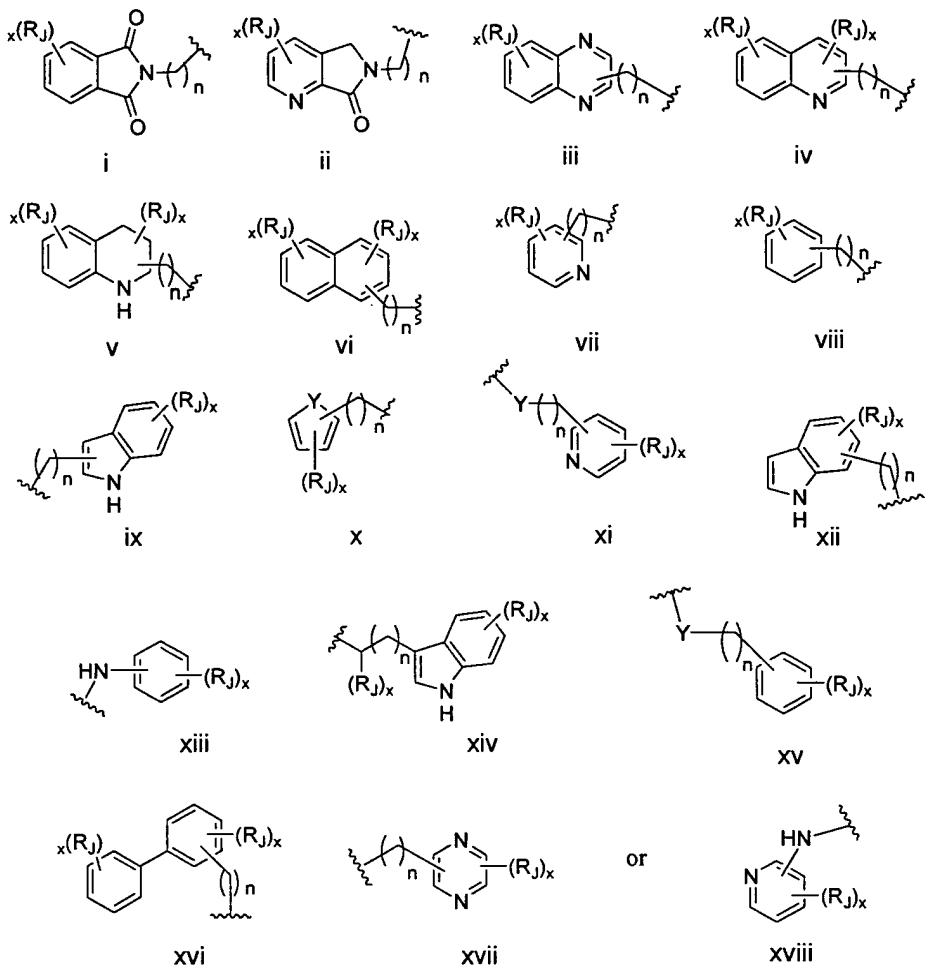
whereby each of said aliphatic and heteroaliphatic moieties is independently substituted or unsubstituted, branched or unbranched, or cyclic or acyclic, and each of said aryl, heteroaryl and heterocyclic moieties is independently substituted or unsubstituted;

5 xxiii) R_1 is a substituted or unsubstituted, branched or unbranched, cyclic or acyclic aliphatic or heteroaliphatic moiety, or a substituted or unsubstituted aryl or heteroaryl moiety;

xxiv) R_1 is an aryl, (aliphatic)aryl, (aliphatic)heteroaryl, heteroaryl, (heteroaliphatic)aryl, or (heteroaliphatic)heteroaryl moiety;

10 whereby each of said aliphatic and heteroaliphatic moieties is independently substituted or unsubstituted, branched or unbranched, or cyclic or acyclic, and each of said aryl, heteroaryl and heterocyclic moieties is independently substituted or unsubstituted;

xxv) any one or more of R_1 , R_A , R_B , R_C , or R_D is independently any one of the following groups:

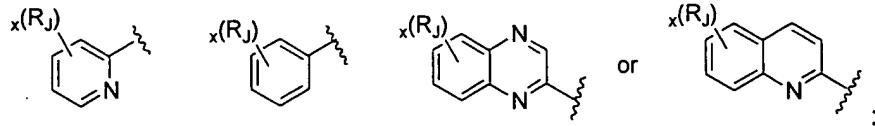


wherein each occurrence of R_J is independently hydrogen, a protecting group, $-OR_K$, $=O$, $-C(=O)R_K$, $-CO_2R_K$, $-CN$, $-SCN$, halogen, $-SR_K$, $-SOR_K$, $-SO_2R_K$, $-NO_2$, $-N(R_K)_2$, $-NHC(O)R_K$,
 5 $-B(OR_K)_2$, or an aliphatic, heteroaliphatic, aryl, or heteroaryl moiety, wherein each occurrence of R_K is independently hydrogen, or an aliphatic, heteroaliphatic, aryl, or heteroaryl moiety, or
 wherein two occurrences of R_K , taken together form a cyclic aliphatic or heteroaliphatic moiety;
 wherein each occurrence of Y is independently O, S or NH; wherein each occurrence of x is
 independently 0-5; and wherein each occurrence of n is independently 0-3, or wherein R_J is a
 10 labeling reagent,

whereby each of said aliphatic and heteroaliphatic moieties are independently substituted or unsubstituted, branched or unbranched or cyclic or acyclic, and each of said aryl and heteroaryl moieties is independently substituted or unsubstituted;

xxvi) R_A is hydrogen, R_B is -(C=O)R_C, and R_C is iii, iv, vii, viii, ix, x, xv, or xvii, or R_A and R_C taken together form the structure of i or ii;

xxvii) R_A is hydrogen, R_B is -(C=O)R_C, and R_C is



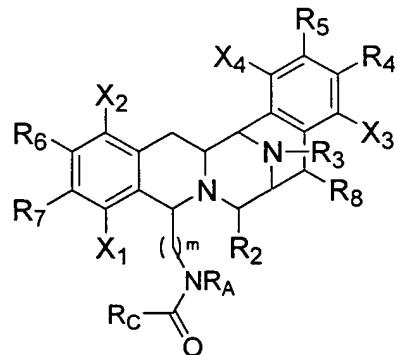
5 xxviii) R_j is hydrogen, halogen, -OH, lower alkyl or lower alkoxy;

xxix) R_j is a linker-biotin or a linker-fluorescein moiety; and

xxx) x is 1 or 2.

As the reader will appreciate, compounds of particular interest include, among others,
10 those which share the attributes of one or more of the foregoing subclasses. Some of those
subclasses are illustrated by the following sorts of compounds:

I) Compounds of the formula:



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as described generally above and in classes and subclasses herein.

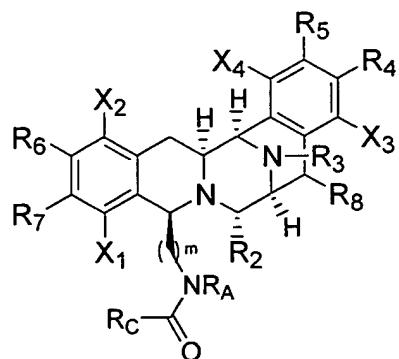
In certain embodiments, for compounds as described directly above, R_A is hydrogen, m is
1 and R_C is an aryl, (aliphatic)aryl, (aliphatic)heteroaryl, heteroaryl, (heteroaliphatic)aryl, or
20 (heteroaliphatic)heteroaryl moiety, or wherein R_A and R_C taken together form a heterocyclic or
heteroaryl moiety, whereby each of said aliphatic and heteroaliphatic moieties is independently
substituted or unsubstituted, branched or unbranched, or cyclic or acyclic, and each of said aryl,
heteroaryl and heterocyclic moieties is independently substituted or unsubstituted.

In certain other embodiments for compounds as described directly above R_A is hydrogen and R_C is any one of groups iii-xviii as described herein, or R_A and R_C taken together are a structure of group i or ii.

In still other embodiments, for compounds as described directly above, X_1 is OH, X_2 is OCH₃, X_3 is OCH₃, X_4 is OH, R_2 is CN, H or OH, R_3 is Me, R_4 is CH₃, R_5 is OCH₃, R_6 is CH₃, R_7 is OCH₃, R_8 is H, R_A is hydrogen, m is 1 and R_C is an aryl, (aliphatic)aryl, (aliphatic)heteroaryl, heteroaryl, (heteroaliphatic)aryl, or (heteroaliphatic)heteroaryl moiety, or wherein R_A and R_C taken together form a heterocyclic or heteroaryl moiety, whereby each of said aliphatic and heteroaliphatic moieties is independently substituted or unsubstituted, 10 branched or unbranched, or cyclic or acyclic, and each of said aryl, heteroaryl and heterocyclic moieties is independently substituted or unsubstituted.

In yet other embodiments, for compounds as described directly above, X_1 is OH, X_2 is OCH₃, X_3 is OCH₃, X_4 is OH, R_2 is CN, H or OH, R_3 is Me, R_4 is CH₃, R_5 is OCH₃, R_6 is CH₃, R_7 is OCH₃, R_8 is H, R_A is hydrogen and R_C is any one of groups iii-xviii as described herein, or 15 R_A and R_C taken together are a structure of group i or ii.

II) Compounds of the formula:



20 as described generally above and in classes and subclasses herein.

In certain embodiments, for compounds as described directly above, R_A is hydrogen, m is 1 and R_C is an aryl, (aliphatic)aryl, (aliphatic)heteroaryl, heteroaryl, (heteroaliphatic)aryl, or (heteroaliphatic)heteroaryl moiety, or wherein R_A and R_C taken together form a heterocyclic or heteroaryl moiety, whereby each of said aliphatic and heteroaliphatic moieties is independently

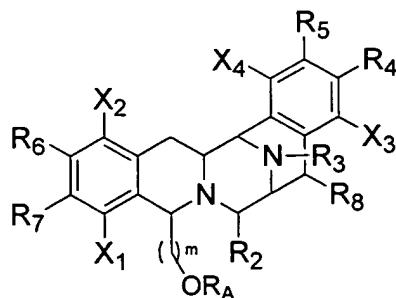
substituted or unsubstituted, branched or unbranched, or cyclic or acyclic, and each of said aryl, heteroaryl and heterocyclic moieties is independently substituted or unsubstituted.

In certain other embodiments for compounds as described directly above R_A is hydrogen and R_C is any one of groups iii-xviii as described herein, or R_A and R_C taken together are a structure of group i or ii.

In still other embodiments, for compounds as described directly above, X_1 is OH, X_2 is OCH_3 , X_3 is OCH_3 , X_4 is OH, R_2 is CN, H or OH, R_3 is Me, R_4 is CH_3 , R_5 is OCH_3 , R_6 is CH_3 , R_7 is OCH_3 , R_8 is H, R_A is hydrogen, m is 1 and R_C is an aryl, (aliphatic)aryl, (aliphatic)heteroaryl, heteroaryl, (heteroaliphatic)aryl, or (heteroaliphatic)heteroaryl moiety, or wherein R_A and R_C taken together form a heterocyclic or heteroaryl moiety, whereby each of said aliphatic and heteroaliphatic moieties is independently substituted or unsubstituted, branched or unbranched, or cyclic or acyclic, and each of said aryl, heteroaryl and heterocyclic moieties is independently substituted or unsubstituted.

In yet other embodiments, for compounds as described directly above, X_1 is OH, X_2 is OCH_3 , X_3 is OCH_3 , X_4 is OH, R_2 is CN, H or OH, R_3 is Me, R_4 is CH_3 , R_5 is OCH_3 , R_6 is CH_3 , R_7 is OCH_3 , R_8 is H, R_A is hydrogen and R_C is any one of groups iii-xviii as described herein, or R_A and R_C taken together are a structure of group i or ii.

III) Compounds of the formula:



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as described generally above and in classes and subclasses herein.

In certain embodiments, for compounds as described directly above, m is 1 and R_A is an aryl, (aliphatic)aryl, (aliphatic)heteroaryl, heteroaryl, (heteroaliphatic)aryl, or (heteroaliphatic)heteroaryl moiety, whereby each of said aliphatic and heteroaliphatic moieties is independently substituted or unsubstituted, branched or unbranched, or cyclic or acyclic, and

each of said aryl, heteroaryl and heterocyclic moieties is independently substituted or unsubstituted.

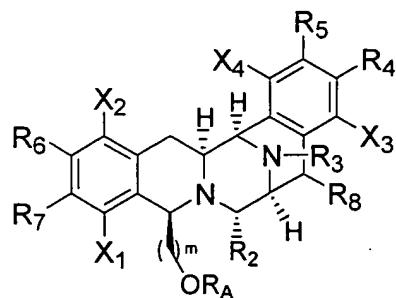
In certain other embodiments for compounds as described directly above R_A is any one of groups iii-xviii as described herein.

5 In still other embodiments, for compounds as described directly above, X₁ is OH, X₂ is OCH₃, X₃ is OCH₃, X₄ is OH, R₂ is CN, H or OH, R₃ is Me, R₄ is CH₃, R₅ is OCH₃, R₆ is CH₃, R₇ is OCH₃, R₈ is H, m is 1 and R_A is an aryl, (aliphatic)aryl, (aliphatic)heteroaryl, heteroaryl, (heteroaliphatic)aryl, or (heteroaliphatic)heteroaryl moiety, whereby each of said aliphatic and heteroaliphatic moieties is independently substituted or unsubstituted, branched or unbranched, 10 or cyclic or acyclic, and each of said aryl, heteroaryl and heterocyclic moieties is independently substituted or unsubstituted.

In yet other embodiments, for compounds as described directly above, X₁ is OH, X₂ is OCH₃, X₃ is OCH₃, X₄ is OH, R₂ is CN, H or OH, R₃ is Me, R₄ is CH₃, R₅ is OCH₃, R₆ is CH₃, R₇ is OCH₃, R₈ is H, R_A is any one of groups iii-xviii as described herein.

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IV) Compounds of the formula:



20 as described generally above and in classes and subclasses herein.

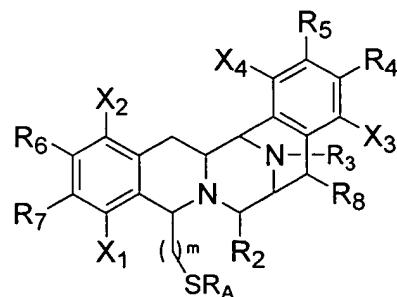
In certain embodiments, for compounds as described directly above, m is 1 and R_A is an aryl, (aliphatic)aryl, (aliphatic)heteroaryl, heteroaryl, (heteroaliphatic)aryl, or (heteroaliphatic)heteroaryl moiety, whereby each of said aliphatic and heteroaliphatic moieties is independently substituted or unsubstituted, branched or unbranched, or cyclic or acyclic, and 25 each of said aryl, heteroaryl and heterocyclic moieties is independently substituted or unsubstituted.

In certain other embodiments for compounds as described directly above R_A is any one of groups iii-xviii as described herein.

In still other embodiments, for compounds as described directly above, X₁ is OH, X₂ is OCH₃, X₃ is OCH₃, X₄ is OH, R₂ is CN, H or OH, R₃ is Me, R₄ is CH₃, R₅ is OCH₃, R₆ is CH₃, 5 R₇ is OCH₃, R₈ is H, m is 1 and R_A is an aryl, (aliphatic)aryl, (aliphatic)heteroaryl, heteroaryl, (heteroaliphatic)aryl, or (heteroaliphatic)heteroaryl moiety, whereby each of said aliphatic and heteroaliphatic moieties is independently substituted or unsubstituted, branched or unbranched, or cyclic or acyclic, and each of said aryl, heteroaryl and heterocyclic moieties is independently substituted or unsubstituted.

10 In yet other embodiments, for compounds as described directly above, X₁ is OH, X₂ is OCH₃, X₃ is OCH₃, X₄ is OH, R₂ is CN, H or OH, R₃ is Me, R₄ is CH₃, R₅ is OCH₃, R₆ is CH₃, R₇ is OCH₃, R₈ is H, R_A is any one of groups iii-xviii as described herein.

V. Compounds of the formula:



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as described generally above and in classes and subclasses herein.

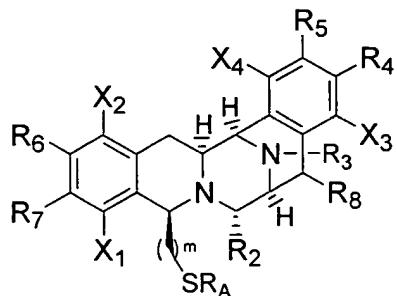
In certain embodiments, for compounds as described directly above, m is 1 and R_A is an aryl, (aliphatic)aryl, (aliphatic)heteroaryl, heteroaryl, (heteroaliphatic)aryl, or (heteroaliphatic)heteroaryl moiety, whereby each of said aliphatic and heteroaliphatic moieties is independently substituted or unsubstituted, branched or unbranched, or cyclic or acyclic, and each of said aryl, heteroaryl and heterocyclic moieties is independently substituted or unsubstituted.

20 In certain other embodiments for compounds as described directly above R_A is any one of groups iii-xviii as described herein.

In still other embodiments, for compounds as described directly above, X₁ is OH, X₂ is OCH₃, X₃ is OCH₃, X₄ is OH, R₂ is CN, H or OH, R₃ is Me, R₄ is CH₃, R₅ is OCH₃, R₆ is CH₃, R₇ is OCH₃, R₈ is H, m is 1 and R_A is an aryl, (aliphatic)aryl, (aliphatic)heteroaryl, heteroaryl, (heteroaliphatic)aryl, or (heteroaliphatic)heteroaryl moiety, whereby each of said aliphatic and
5 heteroaliphatic moieties is independently substituted or unsubstituted, branched or unbranched, or cyclic or acyclic, and each of said aryl, heteroaryl and heterocyclic moieties is independently substituted or unsubstituted.

In yet other embodiments, for compounds as described directly above, X₁ is OH, X₂ is OCH₃, X₃ is OCH₃, X₄ is OH, R₂ is CN, H or OH, R₃ is Me, R₄ is CH₃, R₅ is OCH₃, R₆ is CH₃,
10 R₇ is OCH₃, R₈ is H, R_A is any one of groups iii-xviii as described herein.

VI. Compounds of the formula:



as described generally above and in classes and subclasses herein.

15 In certain embodiments, for compounds as described directly above, m is 1 and R_A is an aryl, (aliphatic)aryl, (aliphatic)heteroaryl, heteroaryl, (heteroaliphatic)aryl, or (heteroaliphatic)heteroaryl moiety, whereby each of said aliphatic and heteroaliphatic moieties is independently substituted or unsubstituted, branched or unbranched, or cyclic or acyclic, and each of said aryl, heteroaryl and heterocyclic moieties is independently substituted or
20 unsubstituted.

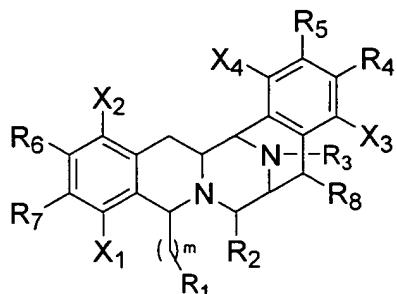
In certain other embodiments for compounds as described directly above R_A is any one of groups iii-xviii as described herein.

In still other embodiments, for compounds as described directly above, X₁ is OH, X₂ is OCH₃, X₃ is OCH₃, X₄ is OH, R₂ is CN, H or OH, R₃ is Me, R₄ is CH₃, R₅ is OCH₃, R₆ is CH₃,
25 R₇ is OCH₃, R₈ is H, m is 1 and R_A is an aryl, (aliphatic)aryl, (aliphatic)heteroaryl, heteroaryl, (heteroaliphatic)aryl, or (heteroaliphatic)heteroaryl moiety, whereby each of said aliphatic and

heteroaliphatic moieties is independently substituted or unsubstituted, branched or unbranched, or cyclic or acyclic, and each of said aryl, heteroaryl and heterocyclic moieties is independently substituted or unsubstituted.

In yet other embodiments, for compounds as described directly above, X₁ is OH, X₂ is OCH₃, X₃ is OCH₃, X₄ is OH, R₂ is CN, H or OH, R₃ is Me, R₄ is CH₃, R₅ is OCH₃, R₆ is CH₃, R₇ is OCH₃, R₈ is H, R_A is any one of groups iii-xviii as described herein.

VII. Compounds of the formula:



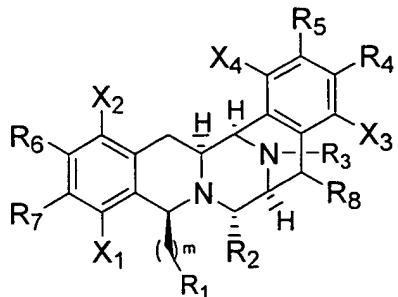
wherein R₁ is an aliphatic, heteroaliphatic, aryl, (aliphatic)aryl, (aliphatic)heteroaryl, heteroaryl, (heteroaliphatic)aryl, or (heteroaliphatic)heteroaryl moiety, whereby each of said aliphatic and heteroaliphatic moieties is independently substituted or unsubstituted, branched or unbranched, or cyclic or acyclic, and each of said aryl, heteroaryl and heterocyclic moieties is independently substituted or unsubstituted.

In certain other embodiments for compounds as described directly above R₁ is any one of groups iii-xviii as described herein.

In still other embodiments, for compounds as described directly above, X₁ is OH, X₂ is OCH₃, X₃ is OCH₃, X₄ is OH, R₂ is CN, H or OH, R₃ is Me, R₄ is CH₃, R₅ is OCH₃, R₆ is CH₃, R₇ is OCH₃, R₈ is H, m is 0 or 1 and R₁ is an aliphatic, heteroaliphatic, aryl, (aliphatic)aryl, (aliphatic)heteroaryl, heteroaryl, (heteroaliphatic)aryl, or (heteroaliphatic)heteroaryl moiety, whereby each of said aliphatic and heteroaliphatic moieties is independently substituted or unsubstituted, branched or unbranched, or cyclic or acyclic, and each of said aryl, heteroaryl and heterocyclic moieties is independently substituted or unsubstituted.

In yet other embodiments, for compounds as described directly above, X₁ is OH, X₂ is OCH₃, X₃ is OCH₃, X₄ is OH, R₂ is CN, H or OH, R₃ is Me, R₄ is CH₃, R₅ is OCH₃, R₆ is CH₃, R₇ is OCH₃, R₈ is H, R₁ is any one of groups iii-xviii as described herein.

VIII. Compounds of the formula:



5 wherein R_1 is an aliphatic, heteroaliphatic, aryl, (aliphatic)aryl, (aliphatic)heteroaryl, heteroaryl, (heteroaliphatic)aryl, or (heteroaliphatic)heteroaryl moiety, whereby each of said aliphatic and heteroaliphatic moieties is independently substituted or unsubstituted, branched or unbranched, or cyclic or acyclic, and each of said aryl, heteroaryl and heterocyclic moieties is independently substituted or unsubstituted.

10 In certain other embodiments for compounds as described directly above R_1 is any one of groups iii-xviii as described herein.

In still other embodiments, for compounds as described directly above, X_1 is OH, X_2 is OCH_3 , X_3 is OCH_3 , X_4 is OH, R_2 is CN, H or OH, R_3 is Me, R_4 is CH_3 , R_5 is OCH_3 , R_6 is CH_3 , R_7 is OCH_3 , R_8 is H, m is 0 or 1 and R_1 is an aliphatic, heteroaliphatic, aryl, (aliphatic)aryl, (aliphatic)heteroaryl, heteroaryl, (heteroaliphatic)aryl, or (heteroaliphatic)heteroaryl moiety, whereby each of said aliphatic and heteroaliphatic moieties is independently substituted or unsubstituted, branched or unbranched, or cyclic or acyclic, and each of said aryl, heteroaryl and heterocyclic moieties is independently substituted or unsubstituted.

15 In yet other embodiments, for compounds as described directly above, X_1 is OH, X_2 is OCH_3 , X_3 is OCH_3 , X_4 is OH, R_2 is CN, H or OH, R_3 is Me, R_4 is CH_3 , R_5 is OCH_3 , R_6 is CH_3 , R_7 is OCH_3 , R_8 is H, R_1 is any one of groups iii-xviii as described herein.

It will be appreciated that some of the foregoing classes and subclasses of compounds can exist in various isomeric forms. The invention encompasses the compounds as individual 20 isomers substantially free of other isomers and alternatively, as mixtures of various isomers, e.g., racemic mixtures of stereoisomers. Additionally, the invention encompasses both (Z) and (E)

double bond isomers unless otherwise specifically designated. The invention also encompasses tautomers of specific compounds as described above. In addition to the above-mentioned compounds per se, this invention also encompasses pharmaceutically acceptable derivatives of these compounds and compositions comprising one or more compounds of the invention and one or more pharmaceutically acceptable excipients or additives.

5 Compounds of this invention which are of particular interest include those which:

- exhibit cytotoxic or growth inhibitory effect on cancer cell lines maintained in vitro or in animal studies using a scientifically acceptable cancer cell xenograft model;
- exhibit enhanced water solubility over existing chemotherapeutic agents, or additionally or alternatively exhibit sufficient solubility to be formulated in an aqueous medium; and
- exhibit a therapeutic profile (e.g., optimum safety and curative effect) that is superior to existing chemotherapeutic agents.

10 15 This invention also provides a pharmaceutical preparation comprising at least one of the compounds as described above and herein, or a pharmaceutically acceptable derivative thereof, which compounds are capable of inhibiting the growth of or killing cancer cells, and, in certain embodiments of special interest are capable of inhibiting the growth of or killing multidrug resistant cancer cells.

20 25 30 The invention further provides a method for inhibiting tumor growth and/or tumor metastasis. In certain embodiments of special interest, the invention provides a method of treating cancers by inhibiting tumor growth and/or tumor metastasis for tumors multidrug resistant cancer cells. The method involves the administration of a therapeutically effective amount of the compound or a pharmaceutically acceptable derivative thereof to a subject (including, but not limited to a human or animal) in need of it. In certain embodiments, specifically for treating cancers comprising multidrug resistant cancer cells, the therapeutically effective amount is an amount sufficient to kill or inhibit the growth of multidrug resistant cancer cell lines. In certain embodiments, the inventive compounds are useful for the treatment of solid tumors.

3) *Compounds and Definitions*

As discussed above, the present invention provides a novel class of compounds useful for the treatment of cancer and other proliferative conditions related thereto. Compounds of this invention comprise those, as set forth above and described herein, and are illustrated in part by 5 the various classes, subgenera and species disclosed elsewhere herein.

It will be appreciated by one of ordinary skill in the art that numerous asymmetric centers exist in the compounds of the present invention. Thus, inventive compounds and pharmaceutical compositions thereof may be in the form of an individual enantiomer, diastereomer or geometric isomer, or may be in the form of a mixture of stereoisomers. Additionally, in certain 10 embodiments, as detailed herein, the method of the present invention provides for the stereoselective synthesis of alkaloids and analogues thereof. Thus, in certain embodiments, the compounds of the invention are enantiopure.

Additionally, the present invention provides pharmaceutically acceptable derivatives of the inventive compounds, and methods of treating a subject using these compounds, 15 pharmaceutical compositions thereof, or either of these in combination with one or more additional therapeutic agents. The phrase, "pharmaceutically acceptable derivative", as used herein, denotes any pharmaceutically acceptable salt, ester, or salt of such ester, of such compound, or any other adduct or derivative which, upon administration to a patient, is capable of providing (directly or indirectly) a compound as otherwise described herein, or a metabolite or 20 residue thereof. Pharmaceutically acceptable derivatives thus include among others pro-drugs. A pro-drug is a derivative of a compound, usually with significantly reduced pharmacological activity, which contains an additional moiety that is susceptible to removal *in vivo* yielding the parent molecule as the pharmacologically active species. An example of a pro-drug is an ester that is cleaved *in vivo* to yield a compound of interest. Pro-drugs of a variety of compounds, and 25 materials and methods for derivatizing the parent compounds to create the pro-drugs, are known and may be adapted to the present invention. Certain exemplary pharmaceutical compositions and pharmaceutically acceptable derivatives will be discussed in more detail herein below.

Certain compounds of the present invention, and definitions of specific functional groups are also described in more detail below. For purposes of this invention, the chemical elements 30 are identified in accordance with the Periodic Table of the Elements, CAS version, Handbook of Chemistry and Physics, 75th Ed., inside cover, and specific functional groups are generally

defined as described therein. Additionally, general principles of organic chemistry, as well as specific functional moieties and reactivity, are described in "Organic Chemistry", Thomas Sorrell, University Science Books, Sausalito: 1999, the entire contents of which are incorporated herein by reference. Furthermore, it will be appreciated by one of ordinary skill in the art that

5 the synthetic methods, as described herein, utilize a variety of protecting groups. By the term "protecting group", as used herein, it is meant that a particular functional moiety, e.g., C, O, S, or N, is temporarily blocked so that a reaction can be carried out selectively at another reactive site in a multifunctional compound. In preferred embodiments, a protecting group reacts selectively in good yield to give a protected substrate that is stable to the projected reactions; the

10 protecting group must be selectively removed in good yield by readily available, preferably nontoxic reagents that do not attack the other functional groups; the protecting group forms an easily separable derivative (more preferably without the generation of new stereogenic centers); and the protecting group has a minimum of additional functionality to avoid further sites of reaction. As detailed herein, oxygen, sulfur, nitrogen and carbon protecting groups may be

15 utilized. Exemplary protecting groups are detailed herein, however, it will be appreciated that the present invention is not intended to be limited to these protecting groups; rather, a variety of additional equivalent protecting groups can be readily identified using the above criteria and utilized in the method of the present invention. Additionally, a variety of protecting groups are described in "Protective Groups in Organic Synthesis" Third Ed. Greene, T.W. and Wuts, P.G.,

20 Eds., John Wiley & Sons, New York: 1999, the entire contents of which are hereby incorporated by reference. Furthermore, a variety of carbon protecting groups are described in Myers, A.; Kung, D.W.; Zhong, B.; Movassaghi, M.; Kwon, S. *J. Am. Chem. Soc.* 1999, 121, 8401-8402, the entire contents of which are hereby incorporated by reference.

It will be appreciated that the compounds, as described herein, may be substituted with

25 any number of substituents or functional moieties. In general, the term "substituted" whether preceded by the term "optionally" or not, and substituents contained in formulas of this invention, refer to the replacement of hydrogen radicals in a given structure with the radical of a specified substituent. When more than one position in any given structure may be substituted with more than one substituent selected from a specified group, the substituent may be either the

30 same or different at every position. As used herein, the term "substituted" is contemplated to include all permissible substituents of organic compounds. In a broad aspect, the permissible

substituents include acyclic and cyclic, branched and unbranched, carbocyclic and heterocyclic, aromatic and nonaromatic substituents of organic compounds. For purposes of this invention, heteroatoms such as nitrogen may have hydrogen substituents and/or any permissible substituents of organic compounds described herein which satisfy the valencies of the 5 heteroatoms. Furthermore, this invention is not intended to be limited in any manner by the permissible substituents of organic compounds. Combinations of substituents and variables envisioned by this invention are preferably those that result in the formation of stable compounds useful in the treatment, for example of proliferative disorders, including, but not limited to cancer. The term "stable", as used herein, preferably refers to compounds which possess 10 stability sufficient to allow manufacture and which maintain the integrity of the compound for a sufficient period of time to be detected and preferably for a sufficient period of time to be useful for the purposes detailed herein.

The term "acyl", as used herein, refers to a carbonyl-containing functionality, e.g., -C(=O)R_x, wherein R_x is an aliphatic, heteroaliphatic, aryl, heteroaryl, (aliphatic)aryl, 15 (heteroaliphatic)aryl, heteroaliphatic(aryl) or heteroaliphatic(heteroaryl) moiety, whereby each of the aliphatic, heteroaliphatic, aryl, or heteroaryl moieties is substituted or unsubstituted, or is a substituted (e.g., hydrogen or aliphatic, heteroaliphatic, aryl, or heteroaryl moieties) oxygen or nitrogen containing functionality (e.g., forming a carboxylic acid, ester, or amide functionality).

The term "aliphatic", as used herein, includes both saturated and unsaturated, straight 20 chain (i.e., unbranched), branched, cyclic, or polycyclic aliphatic hydrocarbons, which are optionally substituted with one or more functional groups. As will be appreciated by one of ordinary skill in the art, "aliphatic" is intended herein to include, but is not limited to, alkyl, alkenyl, alkynyl, cycloalkyl, cycloalkenyl, and cycloalkynyl moieties. Thus, as used herein, the term "alkyl" includes straight, branched and cyclic alkyl groups. An analogous convention 25 applies to other generic terms such as "alkenyl", "alkynyl" and the like. Furthermore, as used herein, the terms "alkyl", "alkenyl", "alkynyl" and the like encompass both substituted and unsubstituted groups. In certain embodiments, as used herein, "lower alkyl" is used to indicate those alkyl groups (cyclic, acyclic, substituted, unsubstituted, branched or unbranched) having 1-6 carbon atoms.

In certain embodiments, the alkyl, alkenyl and alkynyl groups employed in the invention 30 contain 1-20 aliphatic carbon atoms. In certain other embodiments, the alkyl, alkenyl, and

alkynyl groups employed in the invention contain 1-10 aliphatic carbon atoms. In yet other embodiments, the alkyl, alkenyl, and alkynyl groups employed in the invention contain 1-8 aliphatic carbon atoms. In still other embodiments, the alkyl, alkenyl, and alkynyl groups employed in the invention contain 1-6 aliphatic carbon atoms. In yet other embodiments, the
5 alkyl, alkenyl, and alkynyl groups employed in the invention contain 1-4 carbon atoms. Illustrative aliphatic groups thus include, but are not limited to, for example, methyl, ethyl, n-propyl, isopropyl, cyclopropyl, -CH₂-cyclopropyl, allyl, n-butyl, sec-butyl, isobutyl, tert-butyl, cyclobutyl, -CH₂-cyclobutyl, n-pentyl, sec-pentyl, isopentyl, tert-pentyl, cyclopentyl, -CH₂-cyclopentyl, n-hexyl, sec-hexyl, cyclohexyl, -CH₂-cyclohexyl moieties and the like, which again,
10 may bear one or more substituents. Alkenyl groups include, but are not limited to, for example, ethenyl, propenyl, butenyl, 1-methyl-2-buten-1-yl, and the like. Representative alkynyl groups include, but are not limited to, ethynyl, 2-propynyl (propargyl), 1-propynyl and the like.

The term "alkoxy", or "thioalkyl" as used herein refers to an alkyl group, as previously defined, attached to the parent molecular moiety through an oxygen atom or through a sulfur atom. In certain embodiments, the alkyl group contains 1-20 aliphatic carbon atoms. In certain other embodiments, the alkyl group contains 1-10 aliphatic carbon atoms. In yet other embodiments, the alkyl, alkenyl, and alkynyl groups employed in the invention contain 1-8 aliphatic carbon atoms. In still other embodiments, the alkyl group contains 1-6 aliphatic carbon atoms. In yet other embodiments, the alkyl group contains 1-4 aliphatic carbon atoms. Examples
15 of alkoxy, include but are not limited to, methoxy, ethoxy, propoxy, isopropoxy, n-butoxy, tert-butoxy, neopentoxy and n-hexoxy. Examples of thioalkyl include, but are not limited to, methylthio, ethylthio, propylthio, isopropylthio, n-butylthio, and the like.

The term "alkylamino" refers to a group having the structure -NHR' wherein R' is alkyl, as defined herein. In certain embodiments, the alkyl group contains 1-20 aliphatic carbon atoms.
25 In certain other embodiments, the alkyl group contains 1-10 aliphatic carbon atoms. In yet other embodiments, the alkyl, alkenyl, and alkynyl groups employed in the invention contain 1-8 aliphatic carbon atoms. In still other embodiments, the alkyl group contains 1-6 aliphatic carbon atoms. In yet other embodiments, the alkyl group contains 1-4 aliphatic carbon atoms. Examples of alkylamino include, but are not limited to, methylamino, ethylamino, iso-propylamino and the
30 like.

Some examples of substituents of the above-described aliphatic (and other) moieties of compounds of the invention include, but are not limited to aliphatic; heteroaliphatic; aryl; heteroaryl; alkylaryl; alkylheteroaryl; alkoxy; aryloxy; heteroalkoxy; heteroaryloxy; alkylthio; arylthio; heteroalkylthio; heteroarylthio; F; Cl; Br; I; -OH; -NO₂; -CN; -SCN; -CF₃; -CH₂CF₃; -CHCl₂; -CH₂OH; -CH₂CH₂OH; -CH₂NH₂; -CH₂SO₂CH₃; -C(O)R_x; -CO₂(R_x)₂; -OC(O)R_x; -OCO₂R_x; -OCON(R_x)₂; -N(R_x)₂; -S(O)₂R_x; -NR_x(CO)R_x or -B(OR_x)₂, wherein each occurrence of R_x independently includes, but is not limited to, aliphatic, heteroaliphatic, aryl, heteroaryl, alkylaryl, or alkylheteroaryl, or wherein any two of R_x, taken together is a cyclic aliphatic, heteroaliphatic, aryl or heteroaryl moiety, wherein any of the aliphatic, heteroaliphatic, alkylaryl, or alkylheteroaryl substituents described above and herein may be substituted or unsubstituted, branched or unbranched, cyclic or acyclic, and wherein any of the aryl or heteroaryl substituents described above and herein may be substituted or unsubstituted. Additional examples of generally applicable substituents are illustrated by the specific embodiments shown in the Examples that are described herein.

In general, the terms "aryl" and "heteroaryl", as used herein, refer to stable mono- or polycyclic, heterocyclic, polycyclic, and polyheterocyclic unsaturated moieties having preferably 3-14 carbon atoms, each of which may be substituted or unsubstituted. Substituents include, but are not limited to, any of the previously mentioned substituents, i.e., the substituents recited for aliphatic moieties, or for other moieties as disclosed herein, resulting in the formation of a stable compound. In certain embodiments of the present invention, "aryl" refers to a mono- or bicyclic carbocyclic ring system having one or two aromatic rings including, but not limited to, phenyl, naphthyl, tetrahydronaphthyl, indanyl, indenyl and the like. In certain embodiments of the present invention, the term "heteroaryl", as used herein, refers to a cyclic aromatic radical having from five to ten ring atoms of which one ring atom is selected from S, O and N; zero, one or two ring atoms are additional heteroatoms independently selected from S, O and N; and the remaining ring atoms are carbon, the radical being joined to the rest of the molecule via any of the ring atoms, such as, for example, pyridyl, pyrazinyl, pyrimidinyl, pyrrolyl, pyrazolyl, imidazolyl, thiazolyl, oxazolyl, isooxazolyl, thiadiazolyl, oxadiazolyl, thiophenyl, furanyl, quinolinyl, isoquinolinyl, and the like.

It will be appreciated that aryl and heteroaryl groups (including bicyclic aryl groups) can be unsubstituted or substituted, wherein substitution includes replacement of one, two or three of

the hydrogen atoms thereon independently with any one or more of the following moieties including, but not limited to: aliphatic; heteroaliphatic; aryl; heteroaryl; alkylaryl; alkylheteroaryl; alkoxy; aryloxy; heteroalkoxy; heteroaryloxy; alkylthio; arylthio; heteroalkylthio; heteroarylthio; F; Cl; Br; I; -OH; -NO₂; -CN; -SCN; -CF₃; -CH₂CF₃; -CHCl₂; -CH₂OH; -CH₂CH₂OH; -CH₂NH₂; -CH₂SO₂CH₃; -C(O)R_x; -CO₂(R_x); -CON(R_x)₂; -OC(O)R_x; -OCO₂R_x; -OCON(R_x)₂; -N(R_x)₂; -S(O)₂R_x; -NR_x(CO)R_x or -B(OR_x)₂, wherein each occurrence of R_x independently includes, but is not limited to, aliphatic, heteroaliphatic, aryl, heteroaryl, alkylaryl, or alkylheteroaryl, or wherein any two of R_x, taken together is a cyclic aliphatic, heteroaliphatic, aryl or heteroaryl moiety, wherein any of the aliphatic, heteroaliphatic, alkylaryl, or alkylheteroaryl substituents described above and herein may be substituted or unsubstituted, branched or unbranched, cyclic or acyclic, and wherein any of the aryl or heteroaryl substituents described above and herein may be substituted or unsubstituted. Additional examples of generally applicable substituents are illustrated by the specific embodiments shown in the Examples that are described herein.

The term "cycloalkyl", as used herein, refers specifically to groups having three to seven, preferably three to ten carbon atoms. Suitable cycloalkyls include, but are not limited to cyclopropyl, cyclobutyl, cyclopentyl, cyclohexyl, cycloheptyl and the like, which, as in the case of other aliphatic, heteroaliphatic or heterocyclic moieties, may optionally be substituted with substituents including, but not limited to aliphatic; heteroaliphatic; aryl; heteroaryl; alkylaryl; alkylheteroaryl; alkoxy; aryloxy; heteroalkoxy; heteroaryloxy; alkylthio; arylthio; heteroalkylthio; heteroarylthio; F; Cl; Br; I; -OH; -NO₂; -CN; -SCN; -CF₃; -CH₂CF₃; -CHCl₂; -CH₂OH; -CH₂CH₂OH; -CH₂NH₂; -CH₂SO₂CH₃; -C(O)R_x; -CO₂(R_x); -CON(R_x)₂; -OC(O)R_x; -OCO₂R_x; -OCON(R_x)₂; -N(R_x)₂; -S(O)₂R_x; -NR_x(CO)R_x or -B(OR_x)₂, wherein each occurrence of R_x independently includes, but is not limited to, aliphatic, heteroaliphatic, aryl, heteroaryl, alkylaryl, or alkylheteroaryl, or wherein any two of R_x, taken together is a cyclic aliphatic, heteroaliphatic, aryl or heteroaryl moiety, wherein any of the aliphatic, heteroaliphatic, alkylaryl, or alkylheteroaryl substituents described above and herein may be substituted or unsubstituted, branched or unbranched, cyclic or acyclic, and wherein any of the aryl or heteroaryl substituents described above and herein may be substituted or unsubstituted. Additional examples of generally applicable substituents are illustrated by the specific embodiments shown in the Examples that are described herein.

The term "heteroaliphatic", as used herein, refers to aliphatic moieties that contain one or more oxygen, sulfur, nitrogen, phosphorus or silicon atoms, e.g., in place of carbon atoms. Heteroaliphatic moieties may be branched, unbranched, cyclic or acyclic and include saturated and unsaturated heterocycles such as morpholino, pyrrolidinyl, etc. In certain embodiments, 5 heteroaliphatic moieties are substituted by independent replacement of one or more of the hydrogen atoms thereon with one or more moieties including, but not limited to aliphatic; heteroaliphatic; aryl; heteroaryl; alkylaryl; alkylheteroaryl; alkoxy; aryloxy; heteroalkoxy; heteroaryloxy; alkylthio; arylthio; heteroalkylthio; heteroarylthio; F; Cl; Br; I; -OH; -NO₂; -CN; -SCN; -CF₃; -CH₂CF₃; -CHCl₂; -CH₂OH; -CH₂CH₂OH; -CH₂NH₂; -CH₂SO₂CH₃; -C(O)R_x; -CO₂(R_x); -CON(R_x)₂; -OC(O)R_x; -OCO₂R_x; -OCON(R_x)₂; -N(R_x)₂; -S(O)₂R_x; -NR_x(CO)R_x or -B(OR_x)₂, wherein each occurrence of R_x independently includes, but is not limited to, aliphatic, heteroaliphatic, aryl, heteroaryl, alkylaryl, or alkylheteroaryl, or wherein any two of R_x, taken together 10 is a cyclic aliphatic, heteroaliphatic, aryl or heteroaryl moiety, wherein any of the aliphatic, heteroaliphatic, alkylaryl, or alkylheteroaryl substituents described above and herein may be substituted or unsubstituted, branched or unbranched, cyclic or acyclic, and wherein any 15 of the aryl or heteroaryl substituents described above and herein may be substituted or unsubstituted. Additional examples of generally applicable substituents are illustrated by the specific embodiments shown in the Examples that are described herein.

The terms "halo" and "halogen" as used herein refer to an atom selected from fluorine, 20 chlorine, bromine and iodine.

The term "haloalkyl" denotes an alkyl group, as defined above, having one, two, or three halogen atoms attached thereto and is exemplified by such groups as chloromethyl, bromoethyl, trifluoromethyl, and the like.

The term "heterocycloalkyl" or "heterocycle", as used herein, refers to a non-aromatic 5-, 25 6- or 7- membered ring or a polycyclic group, including, but not limited to a bi- or tri-cyclic group comprising fused six-membered rings having between one and three heteroatoms independently selected from oxygen, sulfur and nitrogen, wherein (i) each 5-membered ring has 0 to 1 double bonds and each 6-membered ring has 0 to 2 double bonds, (ii) the nitrogen and sulfur heteroatoms may be optionally be oxidized, (iii) the nitrogen heteroatom may optionally be quaternized, and (iv) any of the above heterocyclic rings may be fused to a benzene ring. Representative heterocycles include, but are not limited to, pyrrolidinyl, pyrazolinyl,

pyrazolidinyl, imidazolinyl, imidazolidinyl, piperidinyl, piperazinyl, oxazolidinyl, isoxazolidinyl, morpholinyl, thiazolidinyl, isothiazolidinyl, and tetrahydrofuryl. In certain embodiments, a "substituted heterocycloalkyl or heterocycle" group is utilized and as used herein, refers to a heterocycloalkyl or heterocycle group, as defined above, substituted by the independent replacement of one, two or three of the hydrogen atoms thereon with but are not limited to aliphatic; heteroaliphatic; aryl; heteroaryl; alkylaryl; alkylheteroaryl; alkoxy; aryloxy; heteroalkoxy; heteroaryloxy; alkylthio; arylthio; heteroalkylthio; heteroarylthio; F; Cl; Br; I; -OH; -NO₂; -CN; -SCN; -CF₃; -CH₂CF₃; -CHCl₂; -CH₂OH; -CH₂CH₂OH; -CH₂NH₂; -CH₂SO₂CH₃; -C(O)R_x; -CO₂(R_x); -CON(R_x)₂; -OC(O)R_x; -OCO₂R_x; -OCON(R_x)₂; -N(R_x)₂; -S(O)₂R_x; -NR_x(CO)R_x or -B(OR_x)₂, wherein each occurrence of R_x independently includes, but is not limited to, aliphatic, heteroaliphatic, aryl, heteroaryl, alkylaryl, or alkylheteroaryl, or wherein any two of R_x, taken together is a cyclic aliphatic, heteroaliphatic, aryl or heteroaryl moiety, wherein any of the aliphatic, heteroaliphatic, alkylaryl, or alkylheteroaryl substituents described above and herein may be substituted or unsubstituted, branched or unbranched, cyclic or acyclic, and wherein any of the aryl or heteroaryl substituents described above and herein may be substituted or unsubstituted. Additional examples of generally applicable substituents are illustrated by the specific embodiments shown in the Examples that are described herein.

"Labeled": As used herein, the term "labeled" is intended to mean that a compound has at least one element, isotope or chemical compound attached to enable the detection of the compound. It will be appreciated that certain of the inventive compounds are substituted with a labeling reagent, that, as used herein, is intended to mean an element, isotope or chemical compound attached to the inventive compound directly or through a suitable linker (e.g., substituted or unsubstituted, branched or unbranched, cyclic or acyclic, aliphatic, heteroaliphatic, aryl, heteroaryl moiety). Certain exemplary labeling reagents are described in the Exemplification herein; however, it will be appreciated that the present invention is not intended to be limited to these examples. Rather, a variety of labeling reagents can be employed as substituents for the inventive compounds.

4. Synthetic Methodology:

It will be appreciated that each of the compounds as described above and herein can be synthesized according to the pioneering methodology described in more detail herein; however,

the synthesis of these compounds is not limited to the methodology described herein. Rather, any methods available in the art of synthetic organic chemistry can be utilized to provide the inventive compounds, including combinatorial techniques. Significantly, the methodology as described herein (and as also described in Myers, A.G., Plowright, A.T. *J. Am Chem. Soc.* 2001, 5 123, 5114-5115; and Myers, A.G., Kung, D.W. *J. Am. Chem. Soc.* 1999, 121, 10828-10829, the entire contents of which are hereby incorporated by reference (including Supplemental Materials available via the internet at <http://publ.acs.org>)) enables the efficient production of significant quantities of alkaloid structures that can be utilized directly or modified to generate other modified alkaloid structures.

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In general, the pioneering method of the present invention provides a general method for the rapid synthesis of alkaloids comprising (1) providing a desired number of substituted aldehyde precursors; and (2) reacting said desired number of substituted aldehyde precursors under suitable conditions to effect the directed condensation of said desired substituted aldehyde precursors, whereby an alkaloid is generated. It will be appreciated that the alkaloid structures generated may, in certain embodiments, be natural product precursors, and thus the subsequent reaction of said precursors with suitable reagents yields a natural product or derivatives thereof (e.g., saframycins or derivatives thereof). In certain other embodiments, the alkaloid structures generated represent core structures that can subsequently be functionalized to generate a variety 15 of structures of interest for biological testing and therapeutic use. In certain embodiments of special interest, inventive compounds are prepared using solid phase methodologies as described herein, for the efficient synthesis of large numbers of exemplary compounds. In general, in certain exemplary embodiments, the step of reacting said desired number of substituted aldehyde precursors comprises reacting said precursors under conditions to effect the directed 20 condensation of said desired substituted aldehyde precursors, whereby an alkaloid is generated. In certain embodiments, the substituted aldehyde precursors are α -amino aldehyde precursors. In certain other embodiments, any combination of α -amino aldehyde and other substituted aldehyde precursors may be utilized (see, e.g., Exemplification, Section IV) to diversify the core 25 pentacyclic structures.

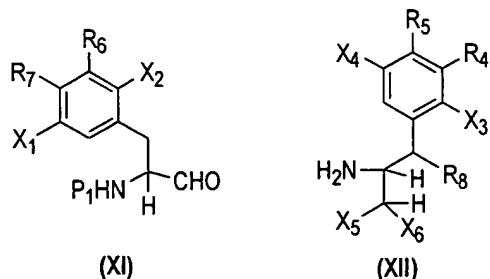
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In certain other embodiments of special interest, particularly those related to the synthesis of saframycins and derivatives thereof, the step of providing said desired number of precursors comprises providing a first N-protected precursor, a second C-protected precursor and a third substituted aldehyde precursor, and the step of reacting said precursors under suitable conditions further comprises: (1) reacting said first N-protected and said second C-protected aldehyde precursor under suitable conditions to generate a tetrahydroisoquinoline core structure; (2) reacting said third substituted aldehyde precursor with said tetrahydroisoquinoline core structure under suitable conditions to generate a trimer of aldehydes; (3) reacting said trimer of aldehydes under suitable conditions to generate an alkaloid; and (4) optionally further reacting the alkaloid generated in step (3) to generate a diversified alkaloid. Additionally, in certain embodiments, the tetrahydroisoquinoline core structure generated in step (1) can be diversified prior to reaction in step (2) with the third aldehyde precursor to generate additional diversity. In certain embodiments, as detailed herein, the third substituted aldehyde precursor is an N-protected α -amino aldehyde precursor. In certain other embodiments, the third aldehyde precursor is an alternate substituted aldehyde precursor, as defined generally. For example, the term "substituted aldehyde precursor", as used herein, refers generally to structures of the formula $R_9(CH_2)_mCHO$, wherein m is 0-5 and R_9 is NR_LR_M , $-OR_L$, $-SR_L$, $-C(=O)R_L$, $-C(=S)R_L$, $-S(O)_2R_L$, or an aliphatic, heteroaliphatic, aryl, heteroaryl, (aliphatic)aryl, (aliphatic)heteroaryl, (heteroaliphatic)aryl, or (heteroaliphatic)heteroaryl moiety, wherein each occurrence of R_L and R_M is independently hydrogen, $-(C=O)R_N$, $-NHR_N$, $-(SO_2)R_N$, $-OR_N$, or an aliphatic, heteroaliphatic, aryl, or heteroaryl moiety, or R_L and R_M , when taken together form an aryl, heteroaryl, cycloaliphatic, or cycloheteroaliphatic moiety, wherein each occurrence of R_N is independently hydrogen, $-OR_P$, $-SR_P$, $-NHR_P$, $-(C=O)R_P$, or an aliphatic, heteroaliphatic, aryl, or heteroaryl moiety, wherein each occurrence of R_P is independently hydrogen, a protecting group, or an aliphatic, heteroaliphatic, aryl, heteroaryl, acyl, alkoxy, aryloxy, alkylthio, arylthio, heteroaryloxy, or heteroarylthio moiety. It will be appreciated that N-protected α -amino aldehyde precursors are encompassed by the generic term "substituted aldehyde precursors". Certain exemplary embodiments are discussed in more detail below and in the exemplification herein.

As described in more detail in the Exemplification section herein, the methodology described generally above takes advantage of the ability to generate a series of imine structures and subsequently subject these imines to suitable reaction conditions to effect cyclization reactions, thus generating the desired alkaloid structures. Thus, in one embodiment of special interest, a first N-protected α -amino aldehyde precursor (**XI**) is provided, and reacted with a second C-protected α -amino aldehyde precursor (**XII**) under suitable conditions to generate a tetrahydroisoquinoline core structure (**XIII**), which reaction comprises first reacting said precursors to effect Schiff-base formation and Pictet-Spengler cyclization and generate a tetrahydroisoquinoline core of structure (**XIII**); and optionally reacting the tetrahydroisoquinoline core structure (**XIII**) to further functionalize the core structure at R₃; and wherein the step of reacting said third aldehyde precursor with said tetrahydroisoquinoline core structure comprises reacting under suitable conditions to effect another Pictet-Spengler cyclization to generate a trimer of aldehydes (**XIV**) and reacting said trimer (**XIV**) under suitable conditions to effect cyclization and generate an alkaloid structure (**XV**).

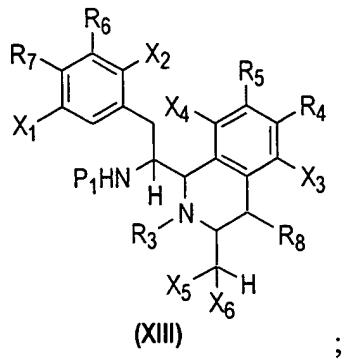
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In certain embodiments of special interest, said first N-protected α -amino aldehyde precursor, and said second C-protected α -amino aldehyde precursor have the general structures:



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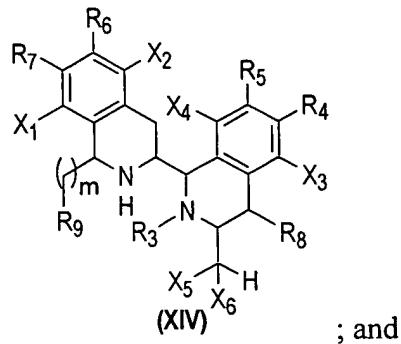
wherein said tetrahydroisoquinoline core has the structure (**XIII**):



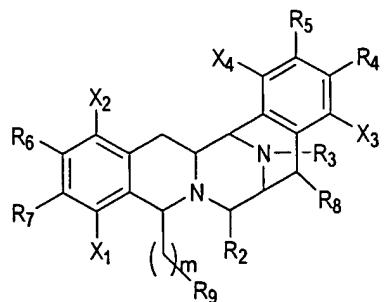
wherein said third aldehyde precursor is $R_9(CH_2)_mCHO$;

wherein said trimer of amino aldehydes has the structure (XIV):

5



wherein the alkaloid generated upon reaction of said trimer under suitable conditions has the structure (XV):



10

(XV)

wherein X_1-X_4 , R_2-R_8 and m are as defined generally and in subclasses herein;

P_1 is hydrogen or a nitrogen protecting group;

X_5 and X_6 taken together represent a carbon protecting group, optionally substituted with

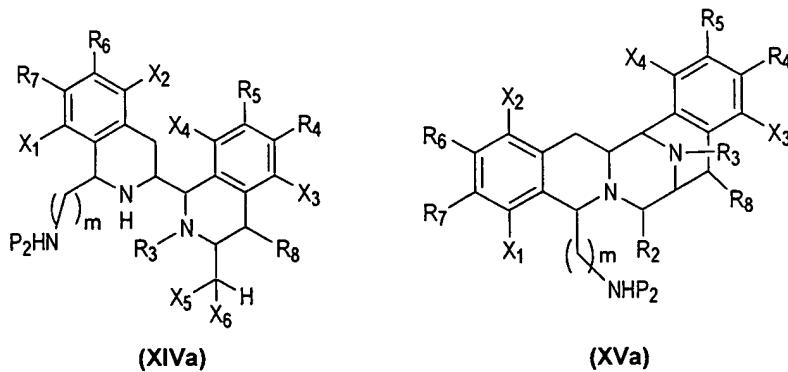
15 a solid support unit; and

R₉ is NR_LR_M, -OR_L, -SR_L, -C(=O)R_L, -C(=S)R_L, -S(O)₂R_L, or an aliphatic, heteroaliphatic, aryl, heteroaryl, (aliphatic)aryl, (aliphatic)heteroaryl, (heteroaliphatic)aryl, or (heteroaliphatic)heteroaryl moiety, wherein each occurrence of R_L and R_M is independently hydrogen, -(C=O)R_N, -NHR_N, -(SO₂)R_N, -OR_N, or an aliphatic, heteroaliphatic, aryl, or heteroaryl moiety, or R_L and R_M, when taken together form an aryl, heteroaryl, cycloaliphatic, or cycloheteroaliphatic moiety, wherein each occurrence of R_N is independently hydrogen, -OR_P, -SR_P, -NHR_P, -(C=O)R_P, or an aliphatic, heteroaliphatic, aryl, or heteroaryl moiety, wherein each occurrence of R_P is independently hydrogen, a protecting group, or an aliphatic, heteroaliphatic, aryl, heteroaryl, acyl, alkoxy, aryloxy, alkylthio, arylthio, heteroaryloxy, or heteroarylthio moiety.

It will be appreciated that, in addition to the synthesis of compounds as described herein, the novel method can be utilized to generate a variety of compounds. For example, the present invention also contemplates the synthesis of ecteinascidin analogues where X₁ and R₇ taken together are a heterocyclic moiety.

In certain embodiments of special interest herein for the intermediates (XIV) and (XV) R₉ is -NHP₂, wherein P₂ is a nitrogen protecting group, and thus the intermediates have the structures (XIVa) and (XVa):

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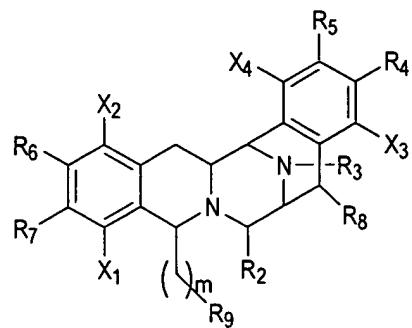
As mentioned above, in certain other embodiments of the invention, compounds of formula (XV) and (XVa) can be further modified to generate compounds of general formula (I) including classes and subclasses thereof, as described in more detail herein.

In certain other embodiments for methods as described herein, the third substituted aldehyde precursor, $R_9(CH_2)_mCHO$ is (aliphatic)(C=O)(CH₂)_mCHO, (heteroaliphatic)(C=O)(CH₂)_mCHO, (aliphatic)(CH₂)_mCHO, (heteroaliphatic)(CH₂)_mCHO, aryl(aliphatic)(CH₂)_mCHO, 5 aryl(heteroaliphatic)(CH₂)_mCHO, -heteroaryl(aliphatic)(CH₂)_mCHO, or heteroaryl(heteroaliphatic)(CH₂)_mCHO, wherein each of the aliphatic, heteroaliphatic, aryl, and heteroaryl moieties is independently substituted or unsubstituted. In certain embodiments, any one or more of the aliphatic, heteroaliphatic, aryl or heteroaryl moieties is substituted with one or 10 more of substituted or unsubstituted amino, substituted or unsubstituted thiol, or substituted or unsubstituted hydroxyl. In certain exemplary embodiments, as described in more detail in the exemplification herein, the third substituted aldehyde precursor is $CH_3(CH_2)_{1-6}CHO$; (protecting group)O(CH₂)₁₋₆CHO; (protecting group)NH(CH₂)₁₋₆CHO; (protecting group)S(CH₂)₁₋₆CHO; (alkyl)O(C=O)CHO; (aryl)(alkenyl)CHO; (heteroaryl)(alkenyl)CHO; (aryl)CHO; or 15 (heteroaryl)CHO, wherein any one or more of the aryl, heteroaryl, alkenyl or alkyl moieties is substituted or unsubstituted.

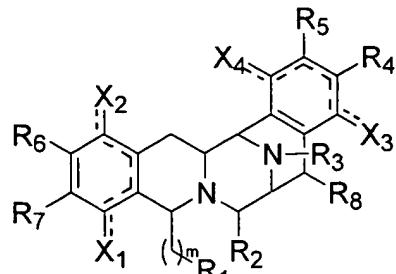
In still other exemplary embodiments as described herein X_5 is CN and X_6 is a heterocyclic moiety optionally substituted with a solid support unit.

20 Thus, the present invention additionally provides a method for the synthesis of compounds of structure (I) as described above and in classes and subclasses herein, which method comprises:

(a) providing a compound of formula (XV)



(b) reacting said compound of formula (XV) under suitable conditions to generate a compound of formula (I):

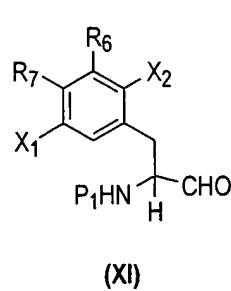


(I)

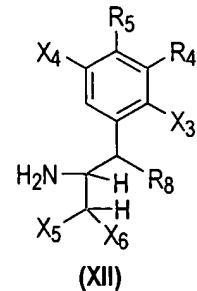
5 wherein X₁-X₄, R₁-R₈, and m are as described above and in classes and subclasses herein, and

wherein the step of providing a compound of formula (XV) further comprises:

10 (1) reacting a first N-protected and a second C-protected α -amino aldehyde precursor having the structures:

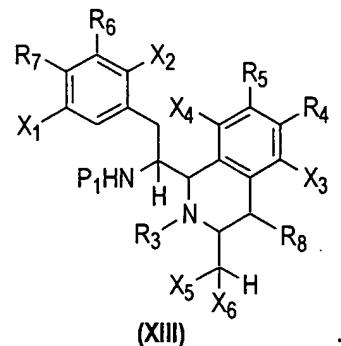


(XI)



(XII)

under suitable conditions to generate a tetrahydroisoquinoline core having the structure (IX):

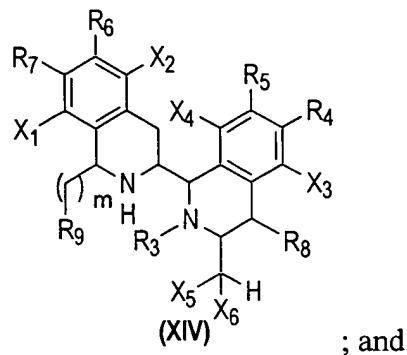


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(2) optionally reacting said tetrahydroisoquinoline core under suitable conditions to diversify R₃;

(3) reacting a third aldehyde precursor having the structure: R₉(CH₂)_mCHO, with said tetrahydroisoquinoline core structure (XIV) under suitable conditions to generate a trimer of aldehydes having the structure:



embodiments, the methods of the present invention enables the stereoselective production of the desired natural product precursors and natural products. In certain embodiments of special interest, the methods as described herein are utilized for the synthesis of an alkaloid structure, wherein the alkaloid structure (**I**) generated is that of saframycin A or derivatives thereof. In
5 certain other particularly preferred embodiments, the method is stereoselective and the alkaloid structure (**I**) generated is that of -(-) saframycin A and derivatives thereof.

It will be appreciated, however, that the method of the present invention can be utilized for the synthesis of any of the compounds as described herein and for classes and subclasses
10 thereof. Additionally, the method of the present invention can be utilized for the synthesis of naturally occurring saframycins and related compounds (e.g., for ecteinascidins and analogues thereof, where R₇ and X₁ taken together are a heterocyclic moiety (methylenedioxy, in certain
embodiments)).

15 It will be appreciated that a variety of experimental conditions can be utilized to effect the synthetic transformations as described generally above. The Exemplification sections describe certain experimental conditions in more detail to enable the production of the compounds of the present invention. It will be appreciated however, that although certain reagents are specifically described in the experimentals, a variety of equivalent reagents can also
20 be utilized. In but one example, although LiBr was utilized to effect activation of the imine functionality to initiate Pictet-Spengler cyclization, it will be appreciated that a variety of other suitable Lewis acids known in the art may be utilized for the activation of the imine functionality as described in the methodology herein. In certain preferred embodiments, those Lewis acids that will not ionise the amino nitrile are utilized in the method of the present invention.
25 Additionally, as described herein, the compounds of the present invention can be diversified at a variety of functional sites (e.g., R₁, R₂, R₃, R₉, oxidation of one or more aromatic rings to quinone moieties, to name a few) either after synthesis of the core structure (**XV**) or during the synthesis of the compounds. As shown in the Exemplification herein, compounds where R₉ is –NHP₁ can be diversified to generate a variety of analogues as described. Additionally, use of a
30 variety of third aldehyde precursors in the method of the present invention yields analogues in which the core ring structure is altered (e.g., -C-C linkages, -C-N linkages, -C-C(=O) linkages

etc.). Additionally, as shown in Figures 14A and 14B, and as described herein, the N-alkylation reaction can be varied to yield exemplary analogues diversified at R₃.

It will be appreciated that the general synthetic method described above can be utilized
5 with solid support techniques. Thus, in certain embodiments of the invention, compounds of the invention are prepared using a solid support. As described herein, the desired alkaloid precursors may be modified or reacted directly to effect attachment to the solid support. The use of a solid support bound component enables the use of more rapid combinatorial (parallel or split-pool) techniques to generate large numbers of compounds more easily. In general, as described in the
10 Exemplification herein, the carbon protected aldehyde can be modified by attachment of a linker moiety (generally an aliphatic or heteroaliphatic moiety) to facilitate attachment of the solid support. The solid support bound carbon protected aldehyde is then reacted under suitable conditions with the first α-amino aldehyde described generally above to generate a solid support bound tetrahydroisoquinoline derivative. Finally, reaction under suitable conditions with a third
15 aldehyde precursor results in the cleavage of the solid support unit and generation of the desired compound.

A solid support, for the purposes of this invention, is defined as an insoluble material to which compounds are attached during a synthesis sequence. The use of a solid support is
20 advantageous for the synthesis of libraries because the isolation of support-bound reaction products can be accomplished simply by washing away reagents from the support-bound material and therefore the reaction can be driven to completion by the use of excess reagents. Additionally, the use of a solid support also enables the use of specific encoding techniques to "track" the identity of the inventive compounds in the library. A solid support can be any
25 material which is an insoluble matrix and can have a rigid or semi-rigid surface. Exemplary solid supports include, but are not limited to, pellets, disks, capillaries, hollow fibers, needles, pins, solid fibers, cellulose beads, pore-glass beads, silica gels, polystyrene beads optionally cross-linked with divinylbenzene, grafted co-poly beads, poly-acrylamide beads, latex beads, dimethylacrylamide beads optionally crosslinked with N-N'-bis-acryloylenediamine, and
30 glass particles coated with a hydrophobic polymer. One of ordinary skill in the art will realize

that the choice of particular solid support will be limited by the compatibility of the support with the reaction chemistry being utilized.

It will be appreciated that specific compounds may be attached directly to the solid
5 support or may be attached to the solid support through a linking reagent. Direct attachment to the solid support may be useful if it is desired not to detach the library member from the solid support. For example, for direct on-bead analysis of biological/pharmacological activity or analysis of the compound structure, a stronger interaction between the library member and the solid support may be desirable. Alternatively, the use of a linking reagent may be useful if more
10 facile cleavage of the inventive library members from the solid support is desired.

Furthermore, any linking reagent used in the present invention may comprise a single linking molecule, or alternatively may comprise a linking molecule and one or more spacer molecules. A spacer molecule is particularly useful when the particular reaction conditions
15 require that the linking molecule be separated from the library member, or if additional distance between the solid support/linking unit and the library member is desired.

Thus, in certain embodiments, libraries of inventive alkaloids can be prepared using established combinatorial methods for solution phase, solid phase, or a combination of solution
20 phase and solid phase synthesis techniques. The synthesis of combinatorial libraries is well known in the art and has been reviewed (see, e.g., "Combinatorial Chemistry", Chemical and Engineering News, Feb. 24, 1997, p. 43; Thompson, L.A., Ellman, J.A., *Chem. Rev.* 1996, 96, 555.) In certain embodiments, the use of solid phase techniques may be desired and thus encoding techniques may also be employed. Specific encoding techniques have been reviewed
25 by Czarnik. (Czarnik, A.W., *Current Opinion in Chemical Biology*, 1997, 1, 60.) One of ordinary skill in the art will realize that the choice of method will depend upon the specific number of compounds to be synthesized, the specific reaction chemistry, and the availability of specific instrumentation, such as robotic instrumentation for the preparation and analysis of libraries. In particularly preferred embodiments, the reactions to be performed on the compound
30 precursors are selected for their ability to proceed in high yield, and in a stereoselective fashion, if desired.

As described in the Exemplification herein, and as shown in Figure 10, in one embodiment of particular interest, the inventive compounds are preparing using a modular solid-supported synthesis. As described generally above, the C-protected aldehyde precursor can be 5 further modified with a solid support unit. As used herein, the term “solid support unit” includes a solid support, as defined herein, and additionally optionally includes a linker moiety which may be an aliphatic, heteroaliphatic, aryl or heteroaryl moiety that facilitates attachment of the solid support to the intermediate of interest. In certain embodiments, herein, a solid support unit is attached to the carbon protecting group (by preparation of a modified group, e.g., preparation 10 of a siloxymorpholine moiety as described in the Exemplification section herein) through a linker unit. It will be appreciated that a variety of linkages and solid supports can be utilized in the method of the invention.

5) *Uses, Formulation and Administration*

15

Pharmaceutical Compositions

As discussed above, the present invention provides novel compounds having antitumor and antiproliferative activity, and thus the inventive compounds are useful for the treatment of cancer. Accordingly, in another aspect of the present invention, pharmaceutical compositions 20 are provided, wherein these compositions comprise any one of the compounds as described herein, and optionally comprise a pharmaceutically acceptable carrier. In certain embodiments, these compositions optionally further comprise one or more additional therapeutic agents. In certain other embodiments, the additional therapeutic agent is an anticancer agent, as discussed in more detail herein.

25 It will also be appreciated that certain of the compounds of present invention can exist in free form for treatment, or where appropriate, as a pharmaceutically acceptable derivative thereof. According to the present invention, a pharmaceutically acceptable derivative includes, but is not limited to, pharmaceutically acceptable salts, esters, salts of such esters, or any other adduct or derivative which upon administration to a patient in need is capable of providing, 30 directly or indirectly, a compound as otherwise described herein, or a metabolite or residue thereof, e.g., a prodrug.

As used herein, the term "pharmaceutically acceptable salt" refers to those salts which are, within the scope of sound medical judgement, suitable for use in contact with the tissues of humans and lower animals without undue toxicity, irritation, allergic response and the like, and are commensurate with a reasonable benefit/risk ratio. Pharmaceutically acceptable salts are well known in the art. For example, S. M. Berge, *et al.* describe pharmaceutically acceptable salts in detail in *J. Pharmaceutical Sciences*, 66: 1-19 (1977), incorporated herein by reference. The salts can be prepared in situ during the final isolation and purification of the compounds of the invention, or separately by reacting the free base function with a suitable organic acid. Examples of pharmaceutically acceptable, nontoxic acid addition salts are salts of an amino group formed with inorganic acids such as hydrochloric acid, hydrobromic acid, phosphoric acid, sulfuric acid and perchloric acid or with organic acids such as acetic acid, oxalic acid, maleic acid, tartaric acid, citric acid, succinic acid or malonic acid or by using other methods used in the art such as ion exchange. Other pharmaceutically acceptable salts include adipate, alginate, ascorbate, aspartate, benzenesulfonate, benzoate, bisulfate, borate, butyrate, camphorate, camphorsulfonate, citrate, cyclopentanepropionate, digluconate, dodecylsulfate, ethanesulfonate, formate, fumarate, glucoheptonate, glycerophosphate, gluconate, hernisulfate, heptanoate, hexanoate, hydroiodide, 2-hydroxy-ethanesulfonate, lactobionate, lactate, laurate, lauryl sulfate, malate, maleate, malonate, methanesulfonate, 2-naphthalenesulfonate, nicotinate, nitrate, oleate, oxalate, palmitate, pamoate, pectinate, persulfate, 3-phenylpropionate, phosphate, picrate, pivalate, propionate, stearate, succinate, sulfate, tartrate, thiocyanate, p-toluenesulfonate, undecanoate, valerate salts, and the like. Representative alkali or alkaline earth metal salts include sodium, lithium, potassium, calcium, magnesium, and the like. Further pharmaceutically acceptable salts include, when appropriate, nontoxic ammonium, quaternary ammonium, and amine cations formed using counterions such as halide, hydroxide, carboxylate, sulfate, phosphate, nitrate, loweralkyl sulfonate and aryl sulfonate.

Additionally, as used herein, the term "pharmaceutically acceptable ester" refers to esters which hydrolyze *in vivo* and include those that break down readily in the human body to leave the parent compound or a salt thereof. Suitable ester groups include, for example, those derived from pharmaceutically acceptable aliphatic carboxylic acids, particularly alkanoic, alkenoic, cycloalkanoic and alkanedioic acids, in which each alkyl or alkenyl moiety advantageously has

not more than 6 carbon atoms. Examples of particular esters include formates, acetates, propionates, butyrates, acrylates and ethylsuccinates.

Furthermore, the term "pharmaceutically acceptable prodrugs" as used herein refers to those prodrugs of the compounds of the present invention which are, within the scope of sound medical judgment, suitable for use in contact with the tissues of humans and lower animals with undue toxicity, irritation, allergic response, and the like, commensurate with a reasonable benefit/risk ratio, and effective for their intended use, as well as the zwitterionic forms, where possible, of the compounds of the invention. The term "prodrug" refers to compounds that are rapidly transformed in vivo to yield the parent compound of the above formula, for example by hydrolysis in blood. A thorough discussion is provided in T. Higuchi and V. Stella, Pro-drugs as Novel Delivery Systems, Vol. 14 of the A.C.S. Symposium Series, and in Edward B. Roche, ed., Bioreversible Carriers in Drug Design, American Pharmaceutical Association and Pergamon Press, 1987, both of which are incorporated herein by reference.

As described above, the pharmaceutical compositions of the present invention additionally comprise a pharmaceutically acceptable carrier, which, as used herein, includes any and all solvents, diluents, or other liquid vehicle, dispersion or suspension aids, surface active agents, isotonic agents, thickening or emulsifying agents, preservatives, solid binders, lubricants and the like, as suited to the particular dosage form desired. Remington's Pharmaceutical Sciences, Sixteenth Edition, E. W. Martin (Mack Publishing Co., Easton, Pa., 1980) discloses various carriers used in formulating pharmaceutical compositions and known techniques for the preparation thereof. Except insofar as any conventional carrier medium is incompatible with the anti-cancer compounds of the invention, such as by producing any undesirable biological effect or otherwise interacting in a deleterious manner with any other component(s) of the pharmaceutical composition, its use is contemplated to be within the scope of this invention. Some examples of materials which can serve as pharmaceutically acceptable carriers include, but are not limited to, sugars such as lactose, glucose and sucrose; starches such as corn starch and potato starch; cellulose and its derivatives such as sodium carboxymethyl cellulose, ethyl cellulose and cellulose acetate; powdered tragacanth; malt; gelatin; talc; excipients such as cocoa butter and suppository waxes; oils such as peanut oil, cottonseed oil; safflower oil; sesame oil; olive oil; corn oil and soybean oil; glycols; such a propylene glycol; esters such as ethyl oleate and ethyl laurate; agar; buffering agents such as magnesium hydroxide and aluminum hydroxide;

alginic acid; pyrogen-free water; isotonic saline; Ringer's solution; ethyl alcohol, and phosphate buffer solutions, as well as other non-toxic compatible lubricants such as sodium lauryl sulfate and magnesium stearate, as well as coloring agents, releasing agents, coating agents, sweetening, flavoring and perfuming agents, preservatives and antioxidants can also be present in the
5 composition, according to the judgment of the formulator.

Uses of Compounds and Pharmaceutical Compositions

In yet another aspect, according to the methods of treatment of the present invention, tumor cells are killed, or their growth is inhibited by contacting said tumor cells with an
10 inventive compound or composition, as described herein. Thus, in still another aspect of the invention, a method for the treatment of cancer is provided comprising administering a therapeutically effective amount of an inventive compound, or a pharmaceutical composition comprising an inventive compound to a subject in need thereof, in such amounts and for such time as is necessary to achieve the desired result. In certain embodiments of the present
15 invention a "therapeutically effective amount" of the inventive compound or pharmaceutical composition is that amount effective for killing or inhibiting the growth of tumor cells. The compounds and compositions, according to the method of the present invention, may be administered using any amount and any route of administration effective for killing or inhibiting the growth of tumor cells. Thus, the expression "amount effective to kill or inhibit the growth of
20 tumor cells", as used herein, refers to a sufficient amount of agent to kill or inhibit the growth of tumor cells. The exact amount required will vary from subject to subject, depending on the species, age, and general condition of the subject, the severity of the infection, the particular anticancer agent, its mode of administration, and the like. The anticancer compounds of the
25 invention are preferably formulated in dosage unit form for ease of administration and uniformity of dosage. The expression "dosage unit form" as used herein refers to a physically discrete unit of anticancer agent appropriate for the patient to be treated. It will be understood, however, that the total daily usage of the compounds and compositions of the present invention will be decided by the attending physician within the scope of sound medical judgment. The specific therapeutically effective dose level for any particular patient or organism will depend
30 upon a variety of factors including the disorder being treated and the severity of the disorder; the activity of the specific compound employed; the specific composition employed; the age, body

weight, general health, sex and diet of the patient; the time of administration, route of administration, and rate of excretion of the specific compound employed; the duration of the treatment; drugs used in combination or coincidental with the specific compound employed; and like factors well known in the medical arts.

5 Furthermore, after formulation with an appropriate pharmaceutically acceptable carrier in a desired dosage, the pharmaceutical compositions of this invention can be administered to humans and other animals orally, rectally, parenterally, intracisternally, intravaginally, intraperitoneally, topically (as by powders, ointments, or drops), buccally, as an oral or nasal spray, or the like, depending on the severity of the infection being treated. In certain
10 embodiments, the compounds of the invention may be administered orally or parenterally at dosage levels of about 0.01 mg/kg to about 50 mg/kg and preferably from about 1 mg/kg to about 25 mg/kg, of subject body weight per day, one or more times a day, to obtain the desired therapeutic effect.

Liquid dosage forms for oral administration include, but are not limited to,
15 pharmaceutically acceptable emulsions, microemulsions, solutions, suspensions, syrups and elixirs. In addition to the active compounds, the liquid dosage forms may contain inert diluents commonly used in the art such as, for example, water or other solvents, solubilizing agents and emulsifiers such as ethyl alcohol, isopropyl alcohol, ethyl carbonate, ethyl acetate, benzyl alcohol, benzyl benzoate, propylene glycol, 1,3-butylene glycol, dimethylformamide, oils (in
20 particular, cottonseed, groundnut, corn, germ, olive, castor, and sesame oils), glycerol, tetrahydrofurfuryl alcohol, polyethylene glycols and fatty acid esters of sorbitan, and mixtures thereof. Besides inert diluents, the oral compositions can also include adjuvants such as wetting agents, emulsifying and suspending agents, sweetening, flavoring, and perfuming agents.

Injectable preparations, for example, sterile injectable aqueous or oleaginous suspensions
25 may be formulated according to the known art using suitable dispersing or wetting agents and suspending agents. The sterile injectable preparation may also be a sterile injectable solution, suspension or emulsion in a nontoxic parenterally acceptable diluent or solvent, for example, as a solution in 1,3-butanediol. Among the acceptable vehicles and solvents that may be employed are water, Ringer's solution, U.S.P. and isotonic sodium chloride solution. In addition, sterile,
30 fixed oils are conventionally employed as a solvent or suspending medium. For this purpose any

bland fixed oil can be employed including synthetic mono- or diglycerides. In addition, fatty acids such as oleic acid are used in the preparation of injectables.

The injectable formulations can be sterilized, for example, by filtration through a bacterial-retaining filter, or by incorporating sterilizing agents in the form of sterile solid compositions which can be dissolved or dispersed in sterile water or other sterile injectable medium prior to use.

In order to prolong the effect of a drug, it is often desirable to slow the absorption of the drug from subcutaneous or intramuscular injection. This may be accomplished by the use of a liquid suspension of crystalline or amorphous material with poor water solubility. The rate of absorption of the drug then depends upon its rate of dissolution which, in turn, may depend upon crystal size and crystalline form. Alternatively, delayed absorption of a parenterally administered drug form is accomplished by dissolving or suspending the drug in an oil vehicle. Injectable depot forms are made by forming microencapsule matrices of the drug in biodegradable polymers such as polylactide-polyglycolide. Depending upon the ratio of drug to polymer and the nature of the particular polymer employed, the rate of drug release can be controlled. Examples of other biodegradable polymers include poly(orthoesters) and poly(anhydrides). Depot injectable formulations are also prepared by entrapping the drug in liposomes or microemulsions which are compatible with body tissues.

Compositions for rectal or vaginal administration are preferably suppositories which can be prepared by mixing the compounds of this invention with suitable non-irritating excipients or carriers such as cocoa butter, polyethylene glycol or a suppository wax which are solid at ambient temperature but liquid at body temperature and therefore melt in the rectum or vaginal cavity and release the active compound.

Solid dosage forms for oral administration include capsules, tablets, pills, powders, and granules. In such solid dosage forms, the active compound is mixed with at least one inert, pharmaceutically acceptable excipient or carrier such as sodium citrate or dicalcium phosphate and/or a) fillers or extenders such as starches, lactose, sucrose, glucose, mannitol, and silicic acid, b) binders such as, for example, carboxymethylcellulose, alginates, gelatin, polyvinylpyrrolidinone, sucrose, and acacia, c) humectants such as glycerol, d) disintegrating agents such as agar--agar, calcium carbonate, potato or tapioca starch, alginic acid, certain silicates, and sodium carbonate, e) solution retarding agents such as paraffin, f) absorption

accelerators such as quaternary ammonium compounds, g) wetting agents such as, for example, cetyl alcohol and glycerol monostearate, h) absorbents such as kaolin and bentonite clay, and i) lubricants such as talc, calcium stearate, magnesium stearate, solid polyethylene glycols, sodium lauryl sulfate, and mixtures thereof. In the case of capsules, tablets and pills, the dosage form
5 may also comprise buffering agents.

Solid compositions of a similar type may also be employed as fillers in soft and hard-filled gelatin capsules using such excipients as lactose or milk sugar as well as high molecular weight polyethylene glycols and the like. The solid dosage forms of tablets, dragees, capsules, pills, and granules can be prepared with coatings and shells such as enteric coatings and other
10 coatings well known in the pharmaceutical formulating art. They may optionally contain opacifying agents and can also be of a composition that they release the active ingredient(s) only, or preferentially, in a certain part of the intestinal tract, optionally, in a delayed manner. Examples of embedding compositions which can be used include polymeric substances and waxes. Solid compositions of a similar type may also be employed as fillers in soft and hard-filled gelatin capsules using such excipients as lactose or milk sugar as well as high molecular
15 weight polyethylene glycols and the like.

The active compounds can also be in micro-encapsulated form with one or more excipients as noted above. The solid dosage forms of tablets, dragees, capsules, pills, and granules can be prepared with coatings and shells such as enteric coatings, release controlling
20 coatings and other coatings well known in the pharmaceutical formulating art. In such solid dosage forms the active compound may be admixed with at least one inert diluent such as sucrose, lactose or starch. Such dosage forms may also comprise, as is normal practice, additional substances other than inert diluents, e.g., tableting lubricants and other tableting aids such as magnesium stearate and microcrystalline cellulose. In the case of capsules, tablets and
25 pills, the dosage forms may also comprise buffering agents. They may optionally contain opacifying agents and can also be of a composition that they release the active ingredient(s) only, or preferentially, in a certain part of the intestinal tract, optionally, in a delayed manner. Examples of embedding compositions which can be used include polymeric substances and waxes.

30 Dosage forms for topical or transdermal administration of a compound of this invention include ointments, pastes, creams, lotions, gels, powders, solutions, sprays, inhalants or patches.

The active component is admixed under sterile conditions with a pharmaceutically acceptable carrier and any needed preservatives or buffers as may be required. Ophthalmic formulation, ear drops, and eye drops are also contemplated as being within the scope of this invention. Additionally, the present invention contemplates the use of transdermal patches, which have the
5 added advantage of providing controlled delivery of a compound to the body. Such dosage forms can be made by dissolving or dispensing the compound in the proper medium. Absorption enhancers can also be used to increase the flux of the compound across the skin. The rate can be controlled by either providing a rate controlling membrane or by dispersing the compound in a polymer matrix or gel.

10 As discussed above, the compounds of the present invention are useful as anticancer agents, and thus may be useful in the treatment of cancer, by effecting tumor cell death or inhibiting the growth of tumor cells. In general, the inventive anticancer agents are useful in the treatment of cancers and other proliferative disorders, including, but not limited to breast cancer, cervical cancer, colon and rectal cancer, leukemia, lung cancer, melanoma, multiple myeloma,
15 non-Hodgkin's lymphoma, ovarian cancer, pancreatic cancer, prostate cancer, and gastric cancer, to name a few. In certain embodiments, the inventive anticancer agents are active against leukemia cells and melanoma cells, and thus are useful for the treatment of leukemias (e.g., myeloid, lymphocytic, myelocytic and lymphoblastic leukemias) and malignant melanomas. In still other embodiments, the inventive anticancer agents are active against solid tumors and also
20 kill and/or inhibit the growth of multidrug resistant cells (MDR cells).

It will also be appreciated that the compounds and pharmaceutical compositions of the present invention can be employed in combination therapies, that is, the compounds and pharmaceutical compositions can be administered concurrently with, prior to, or subsequent to, one or more other desired therapeutics or medical procedures. The particular combination of
25 therapies (therapeutics or procedures) to employ in a combination regimen will take into account compatibility of the desired therapeutics and/or procedures and the desired therapeutic effect to be achieved. It will also be appreciated that the therapies employed may achieve a desired effect for the same disorder (for example, an inventive compound may be administered concurrently with another anticancer agent), or they may achieve different effects (e.g., control of any adverse
30 effects).

For example, other therapies or anticancer agents that may be used in combination with the inventive anticancer agents of the present invention include surgery, radiotherapy (in but a few examples, γ -radiation, neutron beam radiotherapy, electron beam radiotherapy, proton therapy, brachytherapy, and systemic radioactive isotopes, to name a few), endocrine therapy,
5 biologic response modifiers (interferons, interleukins, and tumor necrosis factor (TNF) to name a few), hyperthermia and cryotherapy, agents to attenuate any adverse effects (e.g., antiemetics), and other approved chemotherapeutic drugs, including, but not limited to, alkylating drugs (mechlurethamine, chlorambucil, Cyclophosphamide, Melphalan, Ifosfamide), antimetabolites (Methotrexate), purine antagonists and pyrimidine antagonists (6-Mercaptopurine, 5-
10 Fluorouracil, Cytarabine, Gemcitabine), spindle poisons (Vinblastine, Vincristine, Vinorelbine, Paclitaxel), podophyllotoxins (Etoposide, Irinotecan, Topotecan), antibiotics (Doxorubicin, Bleomycin, Mitomycin), nitrosoureas (Carmustine, Lomustine), inorganic ions (Cisplatin, Carboplatin), enzymes (Asparaginase), and hormones (Tamoxifen, Leuprolide, Flutamide, and Megestrol), to name a few. For a more comprehensive discussion of updated cancer therapies
15 see, <http://www.nci.nih.gov/>, a list of the FDA approved oncology drugs at <http://www.fda.gov/cder/cancer/druglistframe.htm>, and The Merck Manual, Seventeenth Ed. 1999, the entire contents of which are hereby incorporated by reference.

In still another aspect, the present invention also provides a pharmaceutical pack or kit comprising one or more containers filled with one or more of the ingredients of the
20 pharmaceutical compositions of the invention, and in certain embodiments, includes an additional approved therapeutic agent for use as a combination therapy. Optionally associated with such container(s) can be a notice in the form prescribed by a governmental agency regulating the manufacture, use or sale of pharmaceutical products, which notice reflects approval by the agency of manufacture, use or sale for human administration.

25

EQUIVALENTS

The representative examples which follow are intended to help illustrate the invention, and are not intended to, nor should they be construed to, limit the scope of the invention. Indeed, various modifications of the invention and many further embodiments thereof, in addition to
30 those shown and described herein, will become apparent to those skilled in the art from the full contents of this document, including the examples which follow and the references to the

scientific and patent literature cited herein. It should further be appreciated that the contents of those cited references are incorporated herein by reference to help illustrate the state of the art. The following examples contain important additional information, exemplification and guidance which can be adapted to the practice of this invention in its various embodiments and the
5 equivalents thereof.

EXEMPLIFICATION

I. Synthesis of Alkaloids:

A. As discussed above, in one aspect of the invention, novel synthetic methodology
10 for the preparation of alkaloids is provided. In but one embodiment, this methodology involves the directed condensation of α -amino aldehyde precursors. As described in more detail below, the synthesis of saframycin A and intermediates thereof is described.

i) General Description:

Referring to Figures 1 and 2, and Scheme 1, a short and enantioselective synthetic route
15 to the potent antitumor agent (–)-saframycin A (1), a bisquinone alkaloid of microbial origin is provided (for reviews, see, (a) Arai, et al. In *The Alkaloids*; Brossi, A., Ed.; Academic Press: New York, 1983; Vol. 21, Chapter 3 . (b) Remers, W.A. In *The Chemistry of Antitumor Antibiotics*; Wiley-Interscience: New York, 1988; Vol. 2, Chapter 3). The route employs a new and powerful synthetic strategy involving the directed condensation of optically active α -amino
20 aldehydes. This strategy evolved from retrosynthetic analysis of 1, as shown, where a series of transformations initiated by the condensation of an aldehyde with an amine (e.g., reductive amination, Pictet-Spengler, and Strecker reactions), was envisioned to assemble the target 1 from five simple components: hydrogen cyanide, formaldehyde, and three α -amino aldehydes, two of which (structure 3) are the same—hence the latent symmetry of 1. The complexity of the
25 analysis arises in the determination of the precise order and stereochemistry of bonding events that ultimately links the precursors (seven bonds must be formed), and upon consideration of the fundamental issues of stability, reactivity, and protection strategies surrounding the proposed use of optically active α -amino aldehydes as synthetic intermediates. Recently, a series of “C-protected” optically active α -amino aldehydes were reported that incorporate an amino nitrile group as a masked aldehyde (Myers *et al.* *J. Am. Chem. Soc.* **1999**, *121*, 8401). Morpholino nitrile derivatives, exemplified by structure 5, were found to be particularly useful synthetic
30

intermediates, undergoing condensation reactions with optically active *N*-protected α -amino aldehydes with little to no epimerization of either component, thus establishing the basis for the directed assembly of (–)-saframycin A detailed herein.

Compounds **4** and **5**, *N*- and *C*-protected versions of the same chiral α -amino aldehyde (3), were prepared in high enantiomeric excess from the same product of asymmetric alkylation of (–)-pseudoephedrine glycinate, as previously described (Myers *et al.* *J. Am. Chem. Soc.* 1999, 121, 8401; Myers *et al.* *J. Org. Chem.* 1999, 64, 3322). Addition of *N*-protected α -amino aldehyde **4** (96% ee, 1.05 equiv) to *C*-protected α -amino aldehyde **5** (92% ee, 1 equiv) in dichloromethane at 23 °C in the presence of sodium sulfate cleanly provided the imine **6** (presumed trans) without detectable epimerization of either α -stereocenter (^1H NMR analysis, >90% yield, dr ~95:5). Addition of a saturated solution of anhydrous lithium bromide in dimethoxyethane to the imine intermediate and warming to 35 °C brought about Pictet–Spengler cyclization to provide a ~5:1 mixture of cis and trans tetrahydroisoquinolines, respectively. Flash column chromatography afforded the desired cis product (**7**) in 65–72% yield and 99% ee. The optical purity of **7** was assayed by HPLC analysis (Chiralcel OD) of the corresponding bis(benzoyl) derivative against an authentic sample of its enantiomer, derived from (+)-pseudoephedrine via ent-**4** and ent-**5**. Lithium ion proved to be optimal for mild and selective Lewis-acid activation of the imine function without reaction of the morpholino nitrile. The cis-trans selectivity of the cyclization reaction varied markedly as a function of solvent and activating agent; for example, use of lithium perchlorate in diethyl ether provided the trans product exclusively. It is also noteworthy that the transformation of **6** to **7** is the only step in the synthetic route that was conducted above ambient temperature.

Introduction of the *N*-methyl group at this stage of the synthesis was found to be optimal. Stirring **7** at 23 °C in the presence of formalin (2.0 equiv) and sodium triacetoxyborohydride (1.5 equiv) in acetonitrile provided the corresponding *N*-methylated compound in 94% yield; the morpholino nitrile function was unaffected by the reductive conditions. The *N*-Fmoc and OTBS protective groups were then cleaved. While these deprotections could be performed simultaneously by the action of fluoride or hydroxide, sequential removal of the silyl ether with acetic acid-buffered tetrabutylammonium fluoride (2.4 and 1.1 equiv, respectively) followed by cleavage of the carbamate with DBU (1.3 equiv) provided **8** with greater efficiency (92%). Notably, compound **8** showed no propensity for the primary amine to add to the masked

aldehyde under such conditions as exposure to silica gel or upon standing in the protic medium 2,2,2-trifluoroethanol, further highlighting the stability of the morpholino nitrile protective group.

Addition of the third and final α -amino aldehyde component, *N*-Fmoc glycinal (1.5 equiv), to amine **8** (1 equiv) in the presence of sodium sulfate in deoxygenated dichloromethane at 23 °C produced an imine intermediate which underwent Pictet–Spengler cyclization, also at 23 °C, to provide a ≥9:1 mixture of cis and trans tetrahydroisoquinolines, respectively. The desired cis isomer (**9**) was isolated in 66% yield. The reaction solvent was again critical for selective formation of the desired cis tetrahydroisoquinoline; protic solvents afforded predominantly the trans diastereomer (e.g., in methanol, trans:cis >5:1). With the “trimer” of α -amino aldehydes assembled (**9**), the C-terminus morpholino nitrile blocking group was then cleaved with anhydrous zinc chloride (Guibe, F. *et al.* *Tetrahedron Lett.* **1982**, *23*, 5055) (3.0 equiv) in a mixture of trifluoroethanol and tetrahydrofuran (2:1) at 23 °C, producing the key pentacyclic intermediate **2** in 86% yield. This transformation presumably proceeded by the sequential formation of iminium ion **10**, cyclization (addition of the secondary amine to the iminium ion), expulsion of morpholine, and trapping of the resultant iminium ion by cyanide. It was necessary to introduce exogenous cyanide (trimethylsilyl cyanide, 2.0 equiv) to ensure complete amino nitrile formation; in the absence of added cyanide, small amounts (5–10%) of the hemiaminal corresponding to hydrolysis of amino nitrile **2** were observed, presumably due to adventitious water. Finally, the *N*-Fmoc protective group of **2** was cleaved with DBU (1.3 equiv) at 23 °C in 88% yield, and the resulting primary amine was acylated with pyruvyl chloride (3.0 equiv) in the presence of *N,N*-diethylaniline (1.1 equiv) at 0 °C to afford **11** (89%). Oxidative demethylation of the hydroquinones with iodosobenzene (2.5 equiv) in acetonitrile–water (1:1, 0 °C) furnished synthetic (–)-saframycin A in 66% yield (127 mg of (–)-**1**). The synthetic material was found to be identical in all respects (1 H NMR, 13 C NMR, IR, HPLC, tlc analysis, and optical rotation) with an authentic sample of natural saframycin A, kindly provided by Professor T. Arai.

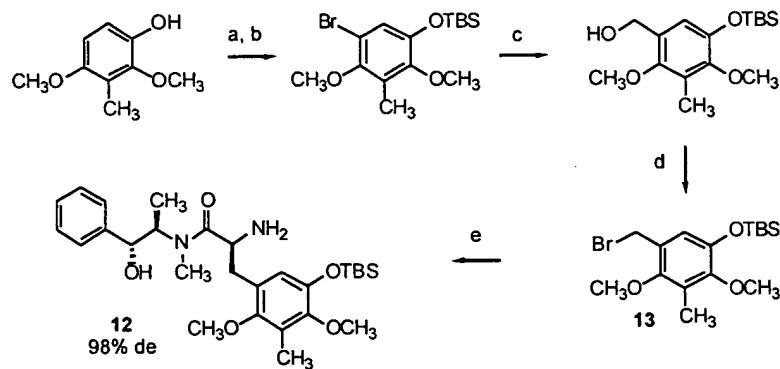
In summary, a practical and efficient synthesis of (–)-saframycin A has been developed that proceeds in just 8 steps from the α -amino aldehyde precursors **4** and **5**, in ~15% overall yield. Significantly, this synthesis illustrates a simple strategy for alkaloid assembly that can be applied more generally, and that involves the directed condensation of α -amino aldehyde precursors in a manner not unlike oligopeptide synthesis (here, with $C\rightarrow N$ directionality). The

present route is suitable for the production of **1** in quantity; to date, more than 1 g of **2** and 200 mg of (-)-**1** have been prepared. Additionally, this synthetic methodology can also be utilized for the generation of **2** in significant quantities and this compound can be further functionalized, as described herein, to generate the compounds as described herein in suitable quantities for therapeutic utility.

ii) Experimental Data:

The *N*- and *C*-protected α -amino aldehyde derivatives **4** and **5** were prepared as previously described (Myers *et al.* *J. Am. Chem. Soc.* **1999**, *121*, 8401) from amide **12**, the product of alkylation (Myers *et al.* *J. Org. Chem.* **1999**, *64*, 3322) of (-)-pseudoephedrine glycinamide with benzylic bromide **13**. Bromide **13** was synthesized from 2,4-dimethoxy-3-methylphenol (Godfrey, I. M.; Sargent, M. V.; Elix, J. A. *J. Chem. Soc., Perkin Trans. 1* **1974**, 1353–1354) as shown in Scheme 1 below.

Scheme 1



(a) TBSCl, imidazole, DMF, 23 °C, 99%. (b) Br₂, pyridine, DMF, 23 °C, 90%. (c) *t*-BuLi, THF, -90 °C; DMF, -90→23 °C; NaBH₄, EtOH, 0 °C, 77%. (d) PPh₃, Br₂, imidazole, CH₂Cl₂, 0 °C, 78%. (e) (-)-pseudoephedrine glycinamide hydrate, LHMDS, LiCl, THF, 0 °C, 74%.

Tetrahydroisoquinoline **7**

A solution of aldehyde **4** (433 mg, 0.752 mmol, 1.05 equiv) in dichloromethane (7.2 mL) was added to a solid mixture of amine **5** (240 mg, 0.716 mmol, 1 equiv) and sodium sulfate (2.03 g, 14.3 mmol, 20.0 equiv). The resulting suspension was stirred rapidly for 75 min at 23 °C,

then was filtered through a plug of cotton. The filtrate was concentrated and the residue was dried azeotropically by concentration from a 3-mL portion of toluene to afford a white foam. ¹H NMR analysis (CDCl₃) showed two diastereomers of imine **6** to be present in a ratio of ~95:5. Anhydrous lithium bromide (1.62 g, 18.7 mmol, 26 equiv) and 1,2-dimethoxyethane (14.3 mL) were then added sequentially to the imine residue, and the mixture was held in a sonicator for 5 min. The resultant suspension was warmed to 35 °C and was held at that temperature for 17.5 h before being cooled to 23 °C. The mixture was diluted with ethyl acetate (20 mL) and was washed with three 20-mL portions of 4:1 saturated aqueous sodium chloride solution–saturated aqueous sodium bicarbonate solution. The organic layer was dried over sodium sulfate and was concentrated. ¹H NMR analysis (CDCl₃) of the residue showed a ~5:1 mixture of cis:trans tetrahydroisoquinolines to be present. The crude product was purified by flash column chromatography (100:1 dichloromethane–methanol) to afford tetrahydroisoquinoline **7** as an off-white solid (460 mg, 72%). HPLC analysis (Chiralcel OD, 4% 2-propanol–0.3% diethylamine–hexanes, 0.30 mL/min, 254 nm detection, ~0.1 mg injection) of bis(benzoyl)-**7** (PhCOCl, Et₃N, DMAP, CH₂Cl₂, 23 °C) established an enantiomeric excess of 99% (t_r(**7**): 79.2 min, t_r(ent-**7**): 89.2 min).

(*R*)-Morpholino nitrile diastereomer, **7**: ¹H NMR (400 MHz, CDCl₃), δ 7.75 (d, 2 H, J = 7.7 Hz, ArH), 7.51 (d, 1 H, J = 8.4 Hz, ArH), 7.44 (d, 1 H, J = 7.3 Hz, ArH), 7.40–7.36 (m, 2 H, ArH), 7.30–7.24 (m, 2 H, ArH), 6.35 (s, 1 H, ArH), 6.20 (s, 1 H, ArOH), 5.70 (d, 1 H, J = 7.0 Hz, NHFmoc), 4.82 (br s, 1 H, ArCHN), 4.58 (m, 1 H, CHNHFmoc), 4.43 (dd, 1 H, J = 9.7, 6.8 Hz, OCH₂CH), 4.19–4.06 (m, 2 H, OCH₂CH), 3.83–3.74 (m, 4 H, CH₂OCH₂), 3.79 (s, 3 H, ArOCH₃), 3.69 (s, 3 H, ArOCH₃), 3.65 (s, 3 H, ArOCH₃), 3.57 (s, 3 H, ArOCH₃), 3.49 (d, 1 H, J = 9.9 Hz, CHC≡N), 3.21 (dd, 1 H, J = 14.6, 2.0 Hz, CH₂ArOH), 3.15–3.08 (m, 1 H, CHCHC≡N), 2.87 (dd, 1 H, J = 13.9, 11.7 Hz, CH₂ArOTBS), 2.73 (m, 2 H, CH₂NCH₂), 2.58 (m, 2 H, CH₂NCH₂), 2.34 (dd, 1 H, J = 14.4, 11.2 Hz, CH₂ArOH), 2.25 (s, 3 H, ArCH₃), 2.19 (s, 3 H, ArCH₃), 2.15 (dd (obsc), 1 H, CH₂ArOTBS), 0.96 (s, 9 H, SiC(CH₃)₃), 0.10 (s, 3 H, SiCH₃), 0.09 (s, 3 H, SiCH₃). ¹³C NMR (100 MHz, CDCl₃), δ 156.4, 151.3, 149.1, 148.6, 145.0, 144.4, 144.2, 142.5, 141.3, 127.6, 127.0, 125.1, 125.0, 124.6, 122.4, 120.2, 119.9, 119.6, 114.7, 66.8, 66.7, 64.1, 60.7, 60.6, 60.4, 59.8, 56.6, 55.0, 50.3, 47.3, 28.9, 28.5, 25.7, 18.2, 9.8, 9.6, –

4.6. FTIR (neat film), cm^{-1} 3345 (w, br, OH/NH), 2934 (s), 2251 (w, C≡N), 1715 (s, C=O), 1471 (s). HRMS (ES) Calcd for $\text{C}_{50}\text{H}_{65}\text{N}_4\text{O}_9\text{Si} (\text{M}+\text{H})^+$: 893.4521, Found: 893.4535.

(S)-Morpholino nitrile diastereomer, 7: ^1H NMR (400 MHz, CDCl_3), δ 7.74 (d, 2 H, $J = 7.3$ Hz, ArH), 7.48 (t, 2 H, $J = 8.0$ Hz, ArH), 7.40–7.35 (m, 2 H, ArH), 7.32–7.24 (m, 2 H, ArH), 6.37 (s, 1 H, ArH), 6.29 (s, 1 H, ArOH), 5.78 (d, 1 H, $J = 7.2$ Hz, NHFmoc), 4.72 (br s, ArCHN), 4.37–4.28 (m, 1 H (CHNHFmoc), 1 H (OCH_2CH)), 4.17–4.11 (m, 2 H, OCH_2CH), 3.78–3.74 (m, 4 H, CH_2OCH_2), 3.76 (s, 3 H, ArOCH₃), 3.67 (s, 6 H, 2 × ArOCH₃), 3.63 (s, 3 H, ArOCH₃), 3.46 (d, 1 H, $J = 7.3$ Hz, CHC□N), 3.24–3.18 (m, 1 H (CH_2ArOH), 1 H (CHCHC□N)), 2.99 (dd, 1 H, $J = 13.9, 10.6$ Hz, CH_2ArOTBS), 2.75–2.71 (m, 2 H, CH_2NCH_2), 2.64–2.60 (m, 2 H, CH_2NCH_2), 2.37–2.28 (m, 1 H (dd, CH_2ArOTBS), 1 H (dd, CH_2ArOH)), 2.22 (s, 3 H, ArCH₃), 2.19 (s, 3 H, ArCH₃), 0.95 (s, 9 H, SiC(CH₃)₃), 0.10 (s, 3 H, SiCH₃), 0.09 (s, 3 H, SiCH₃). ^{13}C NMR (100 MHz, CDCl_3), δ 156.5, 151.4, 148.7, 148.6, 144.9, 144.3, 144.1, 143.9, 142.5, 141.2, 127.6, 127.0, 126.8, 125.2, 125.1, 124.2, 122.4, 120.4, 119.9, 116.1, 66.9, 66.6, 65.3, 60.8, 60.6, 59.8, 57.1, 54.6, 51.3, 50.9, 47.2, 29.2, 27.8, 25.7, 18.2, 9.8, 9.5, –4.6. FTIR (neat film), cm^{-1} 3360 (m, br, OH/NH), 2934 (s), 2249 (w, C≡N), 1715 (s, C=O), 1480 (s). HRMS (ES) Calcd for $\text{C}_{50}\text{H}_{65}\text{N}_4\text{O}_9\text{Si} (\text{M}+\text{H})^+$: 893.4521, Found: 893.4545.

N-Methyl-7

Formalin (554 μL , 7.39 mmol, 2.0 equiv) and sodium triacetoxyborohydride (1.17 g, 5.52 mmol, 1.5 equiv) were added sequentially to a solution of amine 7 (3.30 g, 3.69 mmol, 1 equiv) in acetonitrile (25 mL) at 23 °C. After 30 min, the cloudy mixture was diluted with ethyl acetate (75 mL) and was washed with two 50-mL portions of 1:1 saturated aqueous sodium chloride solution–saturated aqueous sodium bicarbonate solution. The organic layer was dried over sodium sulfate and was concentrated. The residue was purified by flash column chromatography (40% ethyl acetate–hexanes) to afford *N*-methyl-7 as a white solid (3.29 g, 98%).

(R)-Morpholino nitrile diastereomer, *N*-methyl-7: ^1H NMR (400 MHz, toluene- d_8), ~10:1 mixture of atropisomers, major signals only, δ 7.50 (d, 2 H, $J = 7.0$ Hz, ArH), 7.39 (d, 1 H, $J = 6.6$ Hz, ArH), 7.31 (d, 1 H, $J = 7.0$ Hz, ArH), 7.21–7.13 (m, 4 H, ArH), 7.03 (s, 1 H, ArH), 6.09

(br s, 1 H, ArOH), 5.46 (d, 1 H, J = 8.8 Hz, NHFmoc), 4.39–4.32 (m, 1 H, CHNHFmoc), 4.25 (d, 1 H, J = 8.0 Hz, ArCHN), 4.03–3.98 (m, 1 H, OCH₂CH), 3.91–3.84 (m, 2 H, OCH₂CH), 3.58 (s, 3 H, ArOCH₃), 3.54 (s, 3 H, ArOCH₃), 3.46–3.43 (m, 4 H (CH₂OCH₂), 1 H (CHCHC≡N)), 3.44 (s, 3 H, ArOCH₃), 3.35 (dd (obsc), 1 H, CH₂ArOTBS), 3.34 (s, 3 H, ArOCH₃), 3.16–3.10 5 (m, 1 H (d, CHC≡N), 1 H (dd, CH₂ArOTBS)), 3.00 (dd, 1 H, J = 15.0, 12.4 Hz, CH₂ArOH), 2.43–2.35 (m, 2 H (CH₂NCH₂), 1 H (CH₂ArOH)), 2.41 (2 s, 3 H each, ArCH₃ and NCH₃), 2.28 (s, 3 H, ArCH₃), 2.24–2.21 (m, 2 H, CH₂NCH₂), 1.05 (s, 9 H, SiC(CH₃)₃), 0.26 (s, 3 H, SiCH₃), 0.24 (s, 3 H, SiCH₃). ¹³C NMR (100 MHz, CDCl₃), two atropisomers, δ 156.1, 151.5, 148.5, 148.3, 144.8, 144.1, 144.0, 143.9, 142.3, 141.1, 127.5, 127.0, 126.9, 126.7, 125.2, 125.0, 124.9, 10 123.5, 122.6, 120.4, 119.8, 116.0, 66.7, 66.6, 64.6, 63.5, 61.1, 61.0, 60.6, 59.7, 57.5, 52.0, 47.1, 31.1, 25.7, 24.4, 18.1, 9.9, 9.5, –4.6. FTIR (neat film), cm^{–1} 3357 (m, OH/NH), 2936 (s), 2250 (w, C≡N), 1715 (s, C=O), 1480 (s). HRMS (ES) Calcd for C₅₁H₆₇N₄O₉Si (M+H)⁺: 907.4677, Found: 907.4666.

15 (S)-Morpholino nitrile diastereomer, N-methyl-7: ¹H NMR (400 MHz, toluene-d₈), ~7:1 mixture of atropisomers, major signals only, δ 7.49 (d, 2 H, J = 6.9 Hz, ArH), 7.35 (d, 1 H, J = 7.1 Hz, ArH), 7.28 (dd, 1 H, J = 6.3, 2.8 Hz, ArH), 7.23–7.18 (m, 4 H, ArH), 6.88 (s, 1 H, ArH), 5.48 (s, 1 H, ArOH), 4.90 (d, 1 H, J = 9.6 Hz, NHFmoc), 4.24–4.15 (m, 1 H, CHNHFmoc), 4.09 (d, 1 H, J = 10.7 Hz, ArCHN), 3.92 (dd, 1 H, J = 8.2, 5.5 Hz, OCH₂CH), 3.89–3.76 (m, 1 H, 20 CH₂CHNHFmoc), 1 H (CHC≡N), 2 H (OCH₂CH), 3.69 (s, 3 H, ArOCH₃), 3.55 (s, 3 H, ArOCH₃), 3.50–3.48 (m, 4 H, CH₂OCH₂), 3.43 (s, 3 H, ArOCH₃), 3.39 (dd, 1 H, J = 15.4, 6.2 Hz, CH₂CHCHC≡N), 3.19 (s, 3 H, ArOCH₃), 2.76–2.67 (m, 1 H (CH₂CHCHC≡N), 1 H (CHCHC≡N)), 2.64 (s, 3 H, NCH₃), 2.61 (dd, 1 H, J = 13.7, 9.8 Hz, CH₂CHNHFmoc), 2.42–2.32 (m, 4 H, CH₂NCH₂), 2.27 (s, 3 H, ArCH₃), 2.00 (s, 3 H, ArCH₃), 1.06 (s, 9 H, SiC(CH₃)₃), 25 0.23 (s, 3 H, SiCH₃), 0.22 (s, 3 H, SiCH₃). ¹³C NMR (100 MHz, CDCl₃), two atropisomers, δ 155.8, 151.6, 148.8, 148.6, 144.6, 144.1, 144.0, 143.7, 142.7, 141.1, 141.0, 127.6, 127.5, 127.0, 126.9, 126.6, 125.1, 123.3, 122.3, 121.3, 120.6, 119.8, 117.3, 66.8, 66.5, 63.6, 61.2, 61.0, 60.7, 60.0, 59.8, 54.9, 50.2, 49.0, 47.1, 33.5, 25.7, 23.6, 18.1, 9.9, 9.5, –4.6. FTIR (neat film), cm^{–1} 3402 (w, br, OH/NH), 2952 (m), 2252 (w, C≡N), 1713 (s, C=O), 1480 (s). HRMS (ES) Calcd 30 for C₅₁H₆₇N₄O₉Si (M+H)⁺: 907.4677, Found: 907.4713.

Amine 8

Acetic acid (530 μ L, 9.28 mmol, 2.4 equiv) and tetrabutylammonium fluoride (1.0 M in tetrahydrofuran, 4.24 mL, 4.24 mmol, 1.1 equiv) were added sequentially to a solution of *N*-methyl-7 (3.50 g, 3.86 mmol, 1 equiv) in tetrahydrofuran (7.7 mL) at 0 °C. The solution was then warmed to 23 °C. After 1.5 h, the solution was diluted with 25% saturated aqueous sodium bicarbonate solution (25 mL) and was extracted with ether (3 \times 25 mL). The combined organic layers were dried over sodium sulfate and were concentrated to afford *N*-Fmoc-8 as a white foam (3.06 g, >99%). A portion of the residue (1.22 g, 1.54 mmol, 1 equiv) was dissolved in dichloromethane (4.4 mL), and the resulting solution was treated with 1,8-diazabicyclo[5.4.0]undec-7-ene (DBU, 300 μ L, 2.01 mmol, 1.3 equiv) to cleave the *N*-Fmoc group. After 30 min, the reaction solution was loaded directly onto a flash chromatography column. Elution with 25:1 dichloromethane–methanol afforded amine 8 as a solid (845 mg, 96%).

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(*R*)-Morpholino nitrile diastereomer, 8: ^1H NMR (400 MHz, CDCl_3), δ 6.86 (s, 1 H, ArH), 3.84 (s, 3 H, ArOCH₃), 3.81–3.76 (m, 4 H, CH₂OCH₂), 3.76 (s, 3 H, ArOCH₃), 3.72 (d (obsc), 1 H, ArCHNCH₃), 3.70 (s, 3 H, ArOCH₃), 3.66 (d, 1 H, J = 2.7 Hz, CHC≡N), 3.52 (dd, 1 H, J = 13.2, 2.2 Hz, CH₂CHNH₂), 3.33 (dd, 1 H, J = 14.7, 4.4 Hz, CH₂CHCHC≡N), 2.97–2.91 (m, 1 H, CHNH₂), 2.80–2.76 (m, 2 H, CH₂NCH₂), 2.74 (m, 1 H, CHCHC≡N), 2.67–2.60 (m, 2 H, CH₂NCH₂), 2.61 (dd (obsc), 1 H, CH₂CHNH₂), 2.60 (s, 3 H, NCH₃), 2.40 (dd, 1 H, J = 14.7, 12.8 Hz, CH₂CHCHC≡N), 2.23 (s, 3 H, ArCH₃), 2.21 (s, 3 H, ArCH₃). ^{13}C NMR (100 MHz, CDCl_3), δ 150.5, 148.2, 146.8, 145.6, 144.4, 143.9, 128.2, 124.5, 124.2, 123.4, 121.8, 116.4, 114.3, 67.2, 66.7, 64.8, 62.0, 61.2, 60.7, 60.5, 60.2, 52.2, 46.8, 33.6, 23.5, 9.9, 9.5. FTIR (neat film), cm⁻¹ 3364 (m, br, OH/NH), 2938 (s), 2250 (w, C≡N), 1455 (s). HRMS (ES) Calcd for C₃₀H₄₃N₄O₇ (M+H)⁺: 571.3132, Found: 571.3106.

(*S*)-Morpholino nitrile diastereomer, 8: ^1H NMR (400 MHz, CDCl_3), δ 6.68 (s, 1 H, ArH), 3.85 (s, 3 H, ArOCH₃), 3.84–3.73 (m, 4 H (CH₂OCH₂), 1 H (ArCHNCH₃)), 3.78 (s, 3 H, ArOCH₃), 3.75 (s, 3 H, ArOCH₃), 3.69 (dd, 1 H, J = 10.3, 3.0 Hz, CH₂CHNH₂), 3.64 (s, 3 H, ArOCH₃), 3.62 (d, 1 H, J = 9.5 Hz, CHC≡N), 3.31 (dd, 1 H, J = 15.6, 6.4 Hz, CH₂CHCHC≡N), 2.97–2.91

(m, 1 H, CHNH₂), 2.90–2.83 (m, 1 H, CHCHC≡N), 2.78–2.73 (m, 2 H, CH₂NCH₂), 2.64–2.59 (m, 2 H, CH₂NCH₂), 2.59 (s, 3 H, NCH₃), 2.35 (dd, 1 H, J = 13.6, 10.6 Hz, CH₂CHNH₂), 2.24 (s, 3 H, ArCH₃), 2.22 (s, 3 H, ArCH₃), 2.10 (dd, 1 H, J = 15.7, 11.6 Hz, CH₂CHCHC≡N). ¹³C NMR (100 MHz, CDCl₃), δ 150.4, 148.5, 146.1, 145.3, 144.6, 144.2, 128.4, 124.6, 124.2, 123.3, 5 121.1, 117.1, 114.4, 67.2, 66.8, 66.7, 61.2, 61.1, 60.7, 60.5, 60.4, 56.3, 50.5, 49.3, 35.0, 23.7, 9.9, 9.5. FTIR (neat film), cm⁻¹ 3360 (m, br, OH/NH), 2941 (m), 2251 (w, C≡N), 1455 (s). HRMS (ES) Calcd for C₃₀H₄₃N₄O₇ (M+H)⁺: 571.3132, Found: 571.3146.

Tetrahydroisoquinoline 9

10

A deoxygenated (3 freeze-pump-thaw cycles) solution of *N*-Fmoc glycinal (190 mg, 0.675 mmol, 1.2 equiv) in dichloromethane (11.3 mL) was transferred by cannula to a solid mixture of amine **8** (322 mg, 0.564 mmol, 1 equiv) and sodium sulfate (1.20 g, 8.45 mmol, 15 equiv). The resulting suspension was stirred for 17.5 h at 23 °C, then was filtered through a plug of cotton, 15 rinsing with a 6-mL portion of dichloromethane. The filtrate was concentrated and the residue was purified by flash column chromatography (50→70% ethyl acetate–hexanes→ethyl acetate→200:1 ethyl acetate–methanol) to afford tetrahydroisoquinoline **9** as a solid (299 mg, 64%).

20 (*R*)-Morpholino nitrile diastereomer, **9**: ¹H NMR (400 MHz, CDCl₃), signals broadened due to atropisomerism, δ 7.76 (dd, 2 H, J = 7.3, 3.5 Hz, ArH), 7.57 (t, 2 H, J = 7.4 Hz, ArH), 7.31 (m, 2 H, ArH), 7.28 (m, 2 H, ArH), 5.38 (br s, 1 H, NHFmoc), 4.40–4.31 (m, 1 H (CHCH₂NHFmoc), 2 H (OCH₂CH)), 4.23 (m, 1 H, OCH₂CH), 3.90 (m, 1 H, CH₂NHFmoc), 3.84–3.65 (m, 1 H (CH₃NCHCHNH), 4 H (CH₂OCH₂)), 3.76 (s, 3 H, ArOCH₃), 3.74 (s, 3 H, ArOCH₃), 3.67 (s, 3 H, ArOCH₃), 3.65 (s, 3 H, ArOCH₃), 3.58–3.56 (m, 1 H (CHC≡N), 1 H (ArCH₂CHNH)), 3.43 (m, 1 H, CH₂NHFmoc), 3.28 (dd, 1 H, CH₂CHNCH₃), 2.86 (m, 1 H, CH₂CHNCH₃), 2.75–2.71 (m, 2 H, CH₂NCH₂), 2.68 (m, 1 H, ArCH₂CHNH), 2.62 (br s, 3 H, NCH₃), 2.60 (m, 2 H, CH₂NCH₂), 2.39 (m, 1 H (CH₂CHNCH₃), 1 H (ArCH₂CHNH)), 2.21 (s, 6 H, 2 × ArCH₃). ¹³C NMR (100 MHz, CDCl₃), δ 157.1, 149.1, 148.4, 145.5, 144.0, 143.7, 143.3, 141.8, 141.2, 127.6, 25 127.0, 125.9, 125.1, 123.3, 122.7, 122.3, 120.4, 119.9, 116.0, 66.8, 66.6, 65.9, 65.0, 61.1, 60.7,

60.6, 60.5, 58.9, 52.4, 52.2, 51.8, 47.2, 46.4, 26.8, 24.0, 9.6, 9.5. FTIR (neat film), cm^{-1} 3278 (m, OH/NH), 2940 (m), 2249 (w, C≡N), 1705 (s, C=O), 1450 (s). HRMS (ES) Calcd for $\text{C}_{47}\text{H}_{56}\text{N}_5\text{O}_9$ ($\text{M}+\text{H}$)⁺: 834.4078, Found: 834.4106.

5 (S)-Morpholino nitrile diastereomer, **9**: ^1H NMR (500 MHz, CDCl_3), signals broadened due to atropisomerism, δ 7.76 (d, 2 H, $J = 7.0$ Hz, ArH), 7.57 (d, 2 H, $J = 6.5$ Hz, ArH), 7.39 (m, 2 H, ArH), 7.30 (m, 2 H, ArH), 5.44 (br s, 1 H, NHFmoc), 4.42 (dd, 1 H, OCH_2CH), 4.35 (m, 1 H $\text{CHCH}_2\text{NHFmoc}$), 4.28 (t, 1 H, OCH_2CH), 4.22 (dd, 1 H, OCH_2CH), 3.85–3.82 (m, 1 H (CH_2NHFmoc), 1 H ($\text{CH}_2\text{CHNCH}_3$)), 3.75–3.65 (m, 4 H (CH_2OCH_2), 1 H ($\text{CHC}\equiv\text{N}$)), 3.74 (br s, 9 H, 3 × ArOCH₃), 3.65 (s, 3 H, ArOCH₃), 3.37 (m, 1 H, CH_2NHFmoc), 3.32 (dd, 1 H, ArCH₂CHNH), 3.27 (m, 1 H, CH₃NCHCHNH), 2.87 (m, 1 H, CH₂CHNCH₃), 2.74 (m, 1 H, ArCH₂CHNH), 2.70 (m, 2 H, CH₂NCH₂), 2.59 (br s, 3 H, NCH₃), 2.53 (m, 2 H, CH₂NCH₂), 2.36 (dd, 1 H, CH₂CHNCH₃), 2.24 (s, 3 H, ArCH₃), 2.22 (s, 3 H, ArCH₃), 2.13 (dd, 1 H, ArCH₂CHNH). ^{13}C NMR (125 MHz, CDCl_3), δ 157.0, 149.1, 148.6, 145.3, 144.1, 143.9, 143.7, 15 141.8, 141.2, 127.6, 127.0, 125.8, 125.1, 123.3, 122.9, 122.5, 121.9, 120.7, 119.9, 116.3, 66.8, 66.6, 66.1, 62.1, 61.1, 60.7, 60.5, 60.3, 57.0, 52.0, 50.5, 48.9, 47.2, 27.7, 24.0, 9.5, 6.0. FTIR (neat film), cm^{-1} 3274 (m, OH/NH), 2939 (m), 2249 (w, C≡N), 1704 (s, C=O), 1451 (s). HRMS (ES) Calcd for $\text{C}_{47}\text{H}_{56}\text{N}_5\text{O}_9$ ($\text{M}+\text{H}$)⁺: 834.4078, Found: 834.4103.

20 Pentacyclic Intermediate 2

A solution of anhydrous zinc chloride in tetrahydrofuran (0.50 M, 4.26 mL, 2.14 mmol, 3.0 equiv) and trimethylsilyl cyanide (190 μL , 1.42 mmol, 2.0 equiv) were added sequentially to a solution of morpholino nitrile **9** (595 mg, 0.713 mmol, 1 equiv) in 2,2,2-trifluoroethanol (8.5 mL) at 23 °C. After 7 h, an aqueous solution of EDTA (20 mL, 0.20 M (ethylenedinitrilo)tetraacetic acid, disodium salt–0.40 M sodium hydroxide, pH 10) was added, and the resulting mixture was extracted with ethyl acetate (2 × 25 mL). The combined organic layers were washed with a 20-mL portion of 1:1 saturated aqueous sodium chloride solution–saturated aqueous sodium bicarbonate solution, then were dried over sodium sulfate and were concentrated. Purification of the residue by radial chromatography (40%→60% ethyl acetate–hexanes) furnished pentacyclic intermediate **2** as a white solid (464 mg, 87%).

¹H NMR (500 MHz, CDCl₃), ~5:1 mixture of rotamers, major signals only, δ 7.75 (t, 2 H, J = 7.9 Hz, ArH), 7.46–7.39 (m, 4 H, ArH), 7.30 (t, 2 H, J = 7.4 Hz, ArH), 5.63 (s, 1 H, ArOH), 5.54 (s, 1 H, ArOH), 4.54 (t, 1 H, J = 5.8 Hz, NHFmoc), 4.32 (dd, 1 H, J = 10.8, 6.8 Hz, OCH₂CH), 4.22 (dd, 1 H, J = 10.8, 6.3 Hz, OCH₂CH), 4.14 (br s, 1 H, ArCHNCH₃), 4.12 (app t, 1 H, J = 4.4 Hz, CHCH₂NHFmoc), 4.06 (app t, 1 H, J = 6.4 Hz, OCH₂CH), 3.73 (s, 3 H, ArOCH₃), 3.68 (br s, 1 H, CHC≡N), 3.60 (2 s, 3 H each, 2 × ArOCH₃), 3.55 (s, 3 H, ArOCH₃), 3.28 (br d, 1 H, J = 7.6 Hz, CHCHC≡N), 3.24–3.20 (m, 1 H (ArCHCHN), 1 H (ArCHCHCH₂), 1 H (CH₂NHFmoc)), 3.11–3.06 (m, 1 H, CH₂NHFmoc), 2.96 (dd, 1 H, J = 18.5, 7.8 Hz, ArCH₂CHNCH₃), 2.33 (d, 1 H, J = 18.5 Hz, ArCH₂CHNCH₃), 2.31 (s, 3 H, NCH₃), 2.18 (s, 3 H, ArCH₃), 2.11 (s, 3 H, ArCH₃), 1.88 (dd, 1 H, J = 15.7, 12.5 Hz, ArCHCHCH₂). ¹³C NMR (100 MHz, CDCl₃), δ 156.0, 148.2, 144.0, 143.7, 143.6, 143.2, 141.6, 141.2, 127.6, 127.0, 125.0, 124.8, 123.8, 122.3, 122.1, 119.8, 119.7, 118.5, 118.1, 116.8, 65.9, 60.7, 60.6, 59.6, 57.1, 56.6, 56.5, 55.2, 47.2, 45.3, 41.7, 25.7, 21.2, 9.6. FTIR (neat film), cm⁻¹ 3395 (m, br, OH/NH), 2938 (m), 2250 (w, C≡N), 1714 (s, C=O), 1463 (s). HRMS (ES) Calcd for C₄₃H₄₇N₄O₈ (M+H)⁺: 747.3394, Found: 747.3424.

N-Fmoc Cleavage of Pentacyclic Intermediate 2

20 1,8-Diazabicyclo[5.4.0]undec-7-ene (DBU, 140 μL, 0.936 mmol, 1.3 equiv) was added to a solution of pentacyclic intermediate **2** (538 mg, 0.720 mmol, 1 equiv) in dichloromethane (2.9 mL) at 23 °C. After 30 min, the reaction solution was loaded directly onto a flash chromatography column. Elution with 25:1→20:1 dichloromethane–methanol afforded the deprotected product as a white solid (330 mg, 87%).

25 ¹H NMR (500 MHz, CDCl₃), δ 4.16 (br s, 1 H, ArCHNCH₃), 4.03 (br d, 1 H, J = 3.2 Hz, CHCH₂NH₂), 4.00 (d, 1 H, J = 2.1 Hz, CHC≡N), 3.74 (2 s, 3 H each, 2 × ArOCH₃), 3.68 (s, 3 H, ArOCH₃), 3.61 (s, 3 H, ArOCH₃), 3.39 (br d, 1 H, J = 7.6 Hz, CHCHC≡N), 3.24 (app dd, 1 H, J = 11.8, 2.4 Hz, ArCH₂CHCHAR), 3.20 (dd, 1 H, J = 15.7, 2.1 Hz, ArCH₂CHCHAR), 3.07 (dd, 1 H, J = 18.4, 8.0 Hz, CH₂CHCHC≡N), 2.84 (dd, 1 H, J = 13.2, 1.8 Hz, CH₂NH₂), 2.60 (dd, 1 H, J = 18.4, 8.0 Hz, CH₂CHCHC≡N).

= 13.4, 5.9 Hz, CH_2NH_2), 2.43 (d, 1 H, $J = 18.4$ Hz, $\text{CH}_2\text{CHCHC}\equiv\text{N}$), 2.32 (s, 3 H, NCH_3), 2.22 (s, 3 H, ArCH_3), 2.18 (s, 3 H, ArCH_3), 1.83 (dd, 1 H, $J = 15.3, 11.9$ Hz, $\text{ArCH}_2\text{CHCHAR}$). ^{13}C NMR (125 MHz, CDCl_3), δ 148.3, 148.0, 144.6, 143.3, 143.1, 142.6, 124.5, 123.8, 122.5, 122.0, 120.1, 118.2, 117.0, 60.7, 60.6, 60.5, 60.4, 59.9, 59.2, 56.5, 56.3, 55.1, 46.0, 41.7, 25.8, 21.5, 9.5
5 (2 C). FTIR (neat film), cm^{-1} 3356 (w, br, OH/NH), 2936 (m), 2250 (w, $\text{C}\equiv\text{N}$), 1463 (s). HRMS (ES) Calcd for $\text{C}_{28}\text{H}_{37}\text{N}_4\text{O}_6$ ($\text{M}+\text{H}$) $^+$: 525.2713, Found: 525.2690.

Pyruvamide 11

10 N,N -Diethylaniline (34 μL , 0.21 mmol, 1.1 equiv) and pyruvoyl chloride (65 μL , 0.61 mmol, 3.0 equiv) were added sequentially to a solution of the primary amine (107 mg, 0.204 mmol, 1 equiv) in dichloromethane (4.1 mL) at 0 °C. After 30 min, a 10-mL portion of half-saturated aqueous sodium bicarbonate solution was added, and the resulting mixture was extracted with ethyl acetate (10 mL). The organic layer was washed once each with 10-mL portions of half-saturated aqueous sodium bicarbonate solution and 1:1 saturated aqueous sodium chloride solution–saturated aqueous sodium bicarbonate solution, then was dried over sodium sulfate and was concentrated. Flash column chromatography (60% ethyl acetate–hexanes) afforded pyruvamide 11 as a white solid (105 mg, 87%).
15

20 ^1H NMR (400 MHz, CDCl_3), δ 6.44 (t, 1 H, $J = 5.6$ Hz, NHCO), 5.71 (s, 1 H, ArOH), 5.59 (s, 1 H, ArOH), 4.23 (br s, 1 H, CHCH_2NHCO), 4.15 (d, 1 H, $J = 2.1$ Hz, ArCHNCH_3), 4.02 (d, 1 H, $J = 2.4$ Hz, $\text{CHC}\equiv\text{N}$), 3.79 (s, 3 H, ArOCH_3), 3.73 (s, 3 H, ArOCH_3), 3.66 (app dt (obsc), 1 H, CH_2NHCO), 3.65 (s, 3 H, ArOCH_3), 3.58 (s, 3 H, ArOCH_3), 3.41 (br d, 1 H, $J = 7.9$ Hz, $\text{CHCHC}\equiv\text{N}$), 3.29 (app dt, 1 H, $J = 13.6, 4.7$ Hz, CH_2NHCO), 3.24–3.20 (m, 1 H (ArCHCHCH₂Ar)), 1 H (ArCHCHCH₂Ar)), 3.05 (dd, 1 H, $J = 18.5, 8.1$ Hz, $\text{CH}_2\text{CHCHC}\equiv\text{N}$), 2.48 (d, 1 H, $J = 18.5$ Hz, $\text{CH}_2\text{CHCHC}\equiv\text{N}$), 2.30 (s, 3 H, NCH_3), 2.23 (s, 3 H, COCH_3), 2.18 (s, 3 H, ArCH_3), 2.16 (s, 3 H, ArCH_3), 1.92 (dd, 1 H, $J = 16.2, 12.1$ Hz, ArCHCHCH₂). ^{13}C NMR (125 MHz, CDCl_3), δ 196.1, 159.9, 148.4, 148.2, 143.5, 143.3 (2 C), 141.5, 125.0, 123.5, 122.6, 122.4, 118.0, 117.5, 116.4, 60.8, 60.7, 60.4, 59.9, 59.6, 56.5 (2 C), 56.3, 55.0, 41.8, 41.5, 25.8,
25 30 24.3, 21.1, 9.6 (2 C). FTIR (neat film), cm^{-1} 3378 (m, br, OH/NH), 2937 (m), 2251 (w, $\text{C}\equiv\text{N}$),

1720 (m, C=O), 1682 (s, C=O), 1463 (s). HRMS (ES) Calcd for $C_{31}H_{39}N_4O_8$ ($M+H$)⁺: 595.2768, Found: 595.2739.

(-)Saframycin A (1)

5

Iodosobenzene (189 mg, 0.859 mmol, 2.5 equiv) was added to a solution of pyruvamide **11** (203 mg, 0.341 mmol, 1 equiv) in 50% acetonitrile–water (8.0 mL) at 0 °C. After 1 h, the reaction mixture was loaded directly onto a C₁₈-silica flash chromatography column and was eluted with 50% acetonitrile–water to provide synthetic (–)-saframycin A (**1**) as a yellow solid (127 mg, 10 66%). Spectral data and chromatographic properties of synthetic **1** were identical to those of an authentic sample of natural saframycin A.

¹H NMR (500 MHz, CDCl₃), δ 6.67 (br dd, 1 H, J = 8.1, 3.7 Hz, NHCO), 4.06 (br d, 1 H, J = 2.0 Hz, CHN(CH₃)CHCHC≡N), 4.02 (2 s, 3 H, each, 2 × OCH₃), 3.99 (d, 1 H, J = 2.4 Hz, CHC≡N), 15 3.97 (m, 1 H, CHCH₂NH), 3.72 (ddd, 1 H, J = 14.2, 8.8, 1.5 Hz, CH₂NH), 3.43 (br d, 1 H, J = 7.6 Hz, CHCHC≡N), 3.26 (dt, 1 H, J = 14.2, 4.2 Hz, CH₂NH), 3.13 (dt, 1 H, J = 11.4, 2.9 Hz, CHNHC≡N), 2.88 (dd, 1 H, J = 17.7, 2.6 Hz, CH₂CHCHNCH₃), 2.82 (dd, 1 H, J = 21.1, 7.6 Hz, CH₂CHCHC≡N), 2.31 (s, 3 H, NCH₃), 2.25 (s, 3 H, COCH₃), 2.24 (d, 1 H, J = 21.0 Hz, CH₂CHCHC≡N), 1.98 (s, 3 H, CCH₃), 1.92 (s, 3 H, CCH₃), 1.28 (ddd, 1 H, J = 17.7, 11.5, 2.7 Hz, CH₂CHNHC≡N). ¹³C NMR (125 MHz, CDCl₃), δ 196.7, 186.6, 185.3, 182.4, 180.8, 160.2, 155.9, 155.6, 141.5, 141.2, 135.6, 135.5, 129.2, 128.3, 116.6, 61.1, 60.9, 58.2, 56.2, 54.5, 54.2, 53.9, 41.6, 40.6, 25.0, 24.2, 21.5, 8.7 (2 C). FTIR (neat film), cm^{−1} 3407 (w, NH), 2944 (w), 2249 (w, C≡N), 1720 (w), 1682 (m, C=O), 1652 (s, C=O), 1615 (m, C=C), 1447 (m). HRMS (ES) Calcd for $C_{29}H_{31}N_4O_8$ ($M+H$)⁺: 563.2142, Found: 563.2169. [α]_D²³ (synthetic **1**) = −25 4.6°. [α]_D²³ (natural **1**) = −4.0°.

B. In yet another embodiment of the present invention, the general methodology as described is exemplified by the one-step construction of the pentacyclic skeleton of saframycin A (**9**) from a trimeric α-amino aldehyde precursor (**8**), as depicted in Figure 3. Specifically, 30 recognizing that synthetic studies have shown that the potent antitumor alkaloid saframycin A

can be assembled from glycine, alanine, and two molecules of tyrosine (Mikami *et al.* *J. Biol. Chem.* **1985**, *260*, 344; Arai *et al.* *Antimicrob. Agents Chemother.* **1985**, *28*, 5; Arai *et al.* In *The Alkaloids*; Brossi, A., Ed.; Academic Press: New York, 1983; Vol. 21, Chapter 3; Remers, W.A. In *The Chemistry of Antitumor Antibiotics*; Wiley-Interscience: New York, 1988; Vol. 2, 5 Chapter 3.) As demonstrated above (in part A), an efficient synthesis for the generation of alkaloids via the directed condensation of α -amino aldehydes was achieved. Over the course of the five steps described above, the components were linked in a stepwise fashion in a sequence involving two Pictet-Spengler cyclization reactions and an intramolecular Strecker reaction, to form the pentacyclic saframycin A precursor **5** (same as **(2)** depicted in Figure 1; see, Myers, 10 A.G.; Kung, D. W. *J. Am. Chem. Soc.* **1999**, *121*, 10828). In yet another embodiment, as demonstrated herein, alkaloid skeletons, specifically the saframycin skeleton as demonstrated below, can be assembled in one remarkable transformation from an N-linked oligomer of three α -amino aldehyde components **2**, **3** and **4**, (Figure 4) a reaction that suggests for the first time a viable pathway linking saframycin A (**1**) with an oligopeptide precursor and, therefore, a 15 possible biosynthetic route.

The specific oligomer that was targeted initially was the trimeric amino nitrile, **6**, in which **2**, **3**, and **4** are linked by sequential Strecker reactions (see Figure 5). The amino nitrile groups serve to covalently join the three α -amino aldehyde components and were proposed to function later as precursors to electrophilic imine or iminium intermediates that would mediate 20 the three cyclization reactions leading to the saframycin skeleton (For selected examples of the use of amino nitriles as imine/iminium ion precursors in biomimetic systems, see (a) Overman, L.E.; Jacobsen, E. J. *Tetrahedron Lett.* **1982**, *23*, 2741 (b) Ksander *et al.* *Helv. Chim. Acta* **1987**, *70*, 1115; (c) Bonin *et al.* *Org. Synth.* **1992**, *70*, 54). Previously, it had been demonstrated that α -amino aldehydes can be coupled using the Strecker reaction without epimerization of the α -stereocenter (Myers *et al.* *J. Am. Chem. Soc.* **1999**, *121*, 8401). Because amino nitrile formation 25 was anticipated to form two diastereomeric products in each case (of no consequence in later C-C bond forming reactions) ^{13}C labeled cyanide was utilized in the synthesis to facilitate ^{13}C NMR analysis of the products. Also, the sequence was begun with a single diastereomer of the C-protected α -amino aldehyde component **3**, bearing a ^{13}C -label on the cyano group.

30 The order of introduction of α -amino aldehyde components was **3** +**2**, then **4**, representing C- to N-terminus directionality in the synthesis. Mixing **3** (1 equiv, 93% ee, ^{13}C -

labeled cyano group) and its N-protected α -amino aldehyde counterpart **2** (1.05 equiv, 96% ee) in dichloromethane with suspended sodium sulfate led to formation of the corresponding imine, cleanly and without α -epimerization, as previously demonstrated above. In this instance, however, the imine was captured by Strecker reaction with hydrogen cyanide in methanol at 23°
5 C (1.6 equiv acetic acid, 1.5 equiv K¹³CN, Figure 5), whereas in the route described above, the imine was cyclized by warming (35°C) in the presence of lithium bromide. The expected α -amino nitriles **7** (1.1:1 mixture of diastereomers) were obtained in 92% yield after isolation by flash column chromatography (Figure 4). Sequential removal of the silyl ethers (triethylamine trihydrofluoride, 2.5 equiv, CH₃CN, 23°C) and the N-Fmoc group (30% piperidine-CH₂Cl₂,
10 23°C, 76%, two steps) of **7** afforded the fully deprotected "dimer" for coupling with the third component, N-Fmoc glycinal (**4**). Attempted Strecker coupling of these components was complicated by internal cyclization of the glycinaldimine intermediate. Recognizing that such a process provided an aminal product that was functionally equivalent to the trimeric α -amino nitrile originally targeted, the condensation reaction was optimized to form this product
15 (compound **8**, Figure 5). Thus, addition of **4** (1.1 equiv) to a solution of the deprotected dimeric α -amino aldehyde (1 equiv) in dichloromethane at 23°C led to smooth condensation in the absence of hydrogen cyanide to afford a product formulated as the cyclic aminals **8**. These products were not stable to chromatography on silica gel, but ¹H- and ¹³C-NMR analysis showed that they had been formed cleanly (~90% combined yield). Only diastereomers were detected
20 spectroscopically, and these were present in the same ratio as the starting material **7**, suggesting that the cyclic aminal has been formed with a single stereochemistry, tentatively assigned as shown in Figure 5.

Subsequently sequential treatment of **8** with the Lewis acids lithium bromide (dimethoxyethane, reflux) and then zinc chloride (trifluoroethanol-THF, 23°C), and assisted by
25 the fact that **8** was fortuitously well separated chromatographically from all other reaction products, it was possible to isolate the desired pentacyclic saframycin A precursor **9** from the reaction mixture in pure form (4%). With further experimentation, conditions were found to bring about the transformation of **8** to **9** in one step, and in higher yield (Figures 3 and 6); heating a solution of **8** in tetrahydrofuran at reflux in the presence of magnesium bromide etherate (20 equiv) afforded **9** in 8.4 and 9.0% yield in two separate experiments. Importantly, N-acylation of
30 **9** with the enantiomeric Mosher acid chlroides followed by HPLC analysis of the amide products

established that **9** had been formed without racemization (**9** was 99% ee). N-Methylation of **9** with formalin and sodium triacetoxyborohydride in acetonitrile afforded the pentacyclic saframycin A precursor **5**, (as depicted in Figure 4) identical with an authentic sample prepared by the earlier synthetic route (¹H NMR, IR, TLC, and HPLC analysis), except for the anticipated
5 spectroscopic differences attributed to the ¹³C label. Intermediate **5** can be transformed into saframycin A in three steps (50% yield) (see, for example, Figure 2).

The one-step conversion of the N-linked oligomer **8** to the pentacyclic intermediate **9** involves an exceptional number of individual steps. Three cyclization reactions occur, and three of the five stereocenters of saframycin A are established in this step. In theory, each of the five
10 stereogenic centers of the precursor **5** (shown in Figure 4) is epimerizable under the reaction conditions. A single epimerization event may divert the course of reaction from **9**. In that product which is formed, the α -amino aldehyde-derived centers are preserved. Many viable sequences can be envisioned to transform **8** into **9**; the pathway shown in Figure 6 is proposed as
15 that which naturally occurs. In background studies, we have found that aminals have a greater propensity to form imine or iminium ion intermediates under mildly acidic conditions than secondary amino nitriles which, in turn, are more labile than tertiary amino nitriles (Myers *et al.* *J. Am. Chem. Soc.* **1999**, *121*, 8401). For this reason, without wishing to be bound by any particular theory, it is proposed that cleavage of the aminal occurs first, followed by trapping of
20 the resultant imine by Pictet-Spengler cyclization, as depicted in Figure 6. Subsequent ionization of the secondary amino nitrile is proposed to initiate a second Pictet-Spengler cyclization. Finally, ionization of the tertiary amino nitrile group leads to internal Strecker reaction to form
25 the pentacyclic product **9**. It is interesting to note that the ordering of the two Pictet-Spengler reactions in this proposed sequence is opposite to that of our earlier stepwise condensation route. Both Pictet-Spengler cyclizations are believed to proceed with cis selectivity, as observed in the earlier stepwise route.

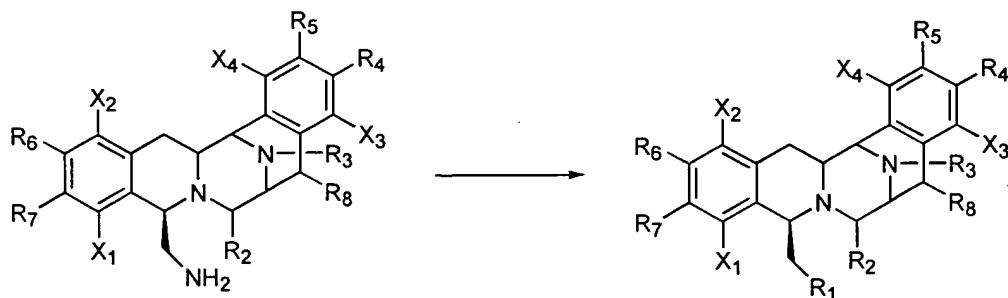
II. Synthesis of analogues of saframycins:

A) **General Procedures:** All reactions were performed in oven-dried or flame-dried round-bottomed flasks. The flasks were fitted with rubber septa or glass stoppers. Commercial
30 reagents were used as received with the following exceptions: THF was distilled from sodium benzophenone ketyl at 760 Torr, methanol was distilled from magnesium methoxide at 760 Torr

and dichloromethane was distilled from calcium hydride at 760 Torr. The spectroscopic and analytical data for all of the analogs is disclosed. All coupling constants are given in Hertz.

2) General Experimentals for analogues:

5 It will be appreciated according to the novel methodology provided by the present invention that useful compounds can be obtained as described herein. In but one example, the methodology of the present invention enables the rapid production of hydroquinones as shown below bearing a protected amino functionality, which amino functionality can be deprotected and reacted with suitable reagents under suitable reaction conditions, certain examples of which are
10 described below, to generate derivatives, as described in more detail herein.



15 In certain embodiments of the present invention, acid chlorides bearing aryl, heteroaryl, aryloxy and alkyl functionalites are reacted with the amino compound presented above under suitable conditions with diethylaniline in methylene chloride to generate desired compounds.

In certain other embodiments of the present invention, aldehydes bearing heteroaryl and aryl functionalities are reacted with the amino compound presented above under suitable conditions with NaBH(OAc)₃ in acetonitrile to generate desired compounds.

20 In still other embodiments of the present invention, carboxylic acids bearing heteroaryl functionalities are reacted with the amino compound presented above under suitable conditions with EDC, HOBT, diethylaniline in tetrahydrofuran to generate desired compounds.

It will be appreciated by one of ordinary skill in the art that a variety of suitable reaction conditions can be utilized to functionalize the amino moiety of the compounds depicted above,
25 and thus the generation of the analogues as described herein is not intended to be limited to the specific examples described herein.

In addition to the syntheses of exemplary analogues from the amino functionality as described generally and depicted above, it will be appreciated that the method of the invention provides for the synthesis of pentacyclic structures bearing (at R₁) O, S and C-containing functionalities, thus enabling access to a variety of analogues.

5 It will be appreciated that the compounds as described herein can be synthesized using traditional solution phase methods (as described generally above), or can be synthesized using solid-support techniques.

3. A Modular, Solid-Supported Synthesis of a Library of (-)-Saframycin A Analogs

10 As described herein, compounds as described generally above and herein can also be prepared using a modular solid-supported synthesis as described herein. Additionally, as described herein, the modular synthesis of the present invention permits extensive diversification of the core structure (e.g., R₁ moieties) as described herein. Furthermore, figures 10-14B depict exemplary schemes and methods for the synthesis of inventive compounds using solid-supported
15 techniques.

Note on Resin Handling:

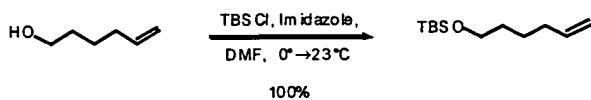
All solid-supported reactions were followed colorimetrically or chromatographically by directly sampling resin beads from the reaction suspension via polypropylene syringe. The
20 sampled beads were then washed on a polypropylene frit (Bio-Spin Disposable Chromatography Columns, P/N: 732-6008, Bio-Rad Laboratories, 2000 Alfred Nobel Drive, Hercules, CA 94547) and dried briefly in vacuo before being employed in colorimetric tests or exposed to cleavage cocktail (Kaiser, E., et. al. *Anal. Biochem.* 1976, 71, 261) (1° amine) and chloranil (Vojkovsky, T. *Pept. Res.*, 1995, 8, 236) (2° amine) colorimetric assays were performed as reported (1999
25 Novabiochem Catalog & Peptide Synthesis Handbook, Calbiochem–Novabiochem Corporation, 10394 Pacific Center Court, San Diego, CA 92121, pp. S43).

The progress of solid-supported reactions was followed chromatographically by liberating product from the solid support by methanolysis of the siloxane linker: Washed resin samples (~5 mg) were suspended in a mixture of 100 µL dichloromethane, 20 µL methanol, and
30 10 µL concentrated hydrochloric acid in a polypropylene Eppendorf tube and allowed to stand for 10 min at 23 °C with occasional manual agitation (~every 3 min). The supernatant from this

reaction mixture was analyzed by thin-layer chromatography, allowing semi-direct monitoring of the transformation of the previously solid-immobilized compounds.

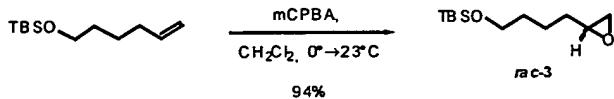
Experimental Procedures:

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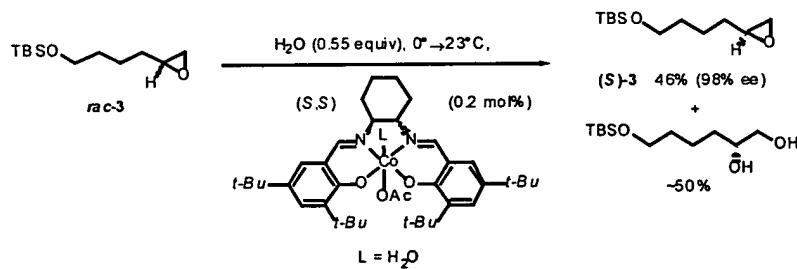
Imidazole (7.33 g, 107.7 mmol, 1.1 equiv) was added in one portion to a solution of 5-hexen-1-ol (9.81 g, 97.91 mmol, 1.0 equiv) in 98.0 mL N,N-dimethylformamide. The resulting clear solution was stirred for 10 min at 0 °C and *t*-butyldimethylsilyl chloride (16.2 g, 107.7 mmol, 1.1 equiv) was added. After stirring an additional 10 min at 0 °C, the resulting reaction solution was allowed to warm to 23 °C and stirred for 1.5 hrs. Excess *t*-butyldimethylsilyl chloride was then quenched by the addition of 100 mL water, and the resulting aqueous solution extracted with 2×200 mL diethyl ether. The combined organic extracts were then washed with 2×400 mL water and 1×500 mL brine, dried over sodium sulfate, and concentrated in vacuo to afford spectroscopically pure siloxane product as a clear oil (22.28 g, 100%).

¹H NMR (500 MHz, CDCl₃), δ 5.85–5.77 (m, 1H, RCHCH₂), 5.00 (dq, 1H, J = 17.5 Hz, RCHCH (Z)), 4.94 (dm, 1H, J = 10.0 Hz, RCHCH (E)), 3.61 (t, 2H, J = 6.5 Hz, CH₂OTBS), 2.06 (q, 2H, J = 6.5 Hz, CH₂CHCH₂), 1.53 (m, 2H, J = 6.0 Hz, CH₂CH₂OTBS), 1.42 (m, 2H, J = 6.0 Hz, CH₂CH₂CHCH₂), 0.90 (s, 9H, SiC(CH₃)₃), 0.02 (s, 6H, Si(CH₃)₂). ¹³C NMR (100 MHz, CDCl₃), δ 139.2, 114.6, 63.3, 33.8, 32.5, 26.2, 25.4, 18.6, -5.1. FTIR (neat film), cm⁻¹ 3079 (w, CH), 2955 (s), 2930 (s), 2858 (s), 1472 (m), 1256 (s), 1103 (s), 960 (m), 836 (s), 775 (s). R_f 0.70, 10% ethyl acetate–hexanes. HRMS (TOF-ES⁺) Calcd for C₁₂H₂₇OSi (M+H)⁺: 215.1831, 25 Found .



3-chloroperoxybenzoic acid (442.0 mg, 77%, 1.97 mmol, 1.2 equiv) was added in one portion to a solution of the siloxane substrate (352.2 mg, 1.64 mmol, 1.0 equiv) in 8.0 mL dichloromethane at 0 °C. After 5 min, the reaction solution was allowed to warm to 23 °C and stir for 13 hr. The reaction solution was then diluted with 80 mL pentane and washed sequentially with 1×80 mL sat. aq. sodium bicarbonate, 1×80 mL sat. aq. sodium bisulfite, 1×80 mL sat. aq. sodium bicarbonate, and 1×80 mL brine. After drying over sodium sulfate, the organic layer was concentrated in vacuo and chromatographically purified (SiO₂, 7% ether–pentane → 20% ether–pentane), providing the epoxide **rac-3** as a clear oil (353.7 mg, 94%).

rac-3: ¹H NMR (400 MHz, CDCl₃), δ 3.62 (t, 2H, J = 6.0 Hz, CH₂OTBS), 2.91 (m, 1H, CH(O)CH₂), 2.75 (dd, 1H, J = 4.0, 5.2 Hz, CH(O)CH₂ (Z)), 2.47 (dd, 1H, J = 2.8, 5.2 Hz, CH(O)CH₂ (E)), 1.61–1.47 (m, 6H, CH₂CH₂CH₂CH₂OTBS), 0.98 (s, 9H, SiC(CH₃)₃), 0.05 (s, 6H, Si(CH₃)₂). ¹³C NMR (100 MHz, CDCl₃), δ 63.1, 52.5, 47.3, 32.8, 32.5, 26.2, 22.6, 18.6, –5.0. FTIR (neat film), cm⁻¹ 2951 (s), 2929 (s), 2856 (s), 1472 (s), 1255 (s), 1099 (s), 836 (s), 775 (s). R_f 0.23, 5% ethyl acetate–hexanes. LRMS (TOF-ES⁺) Calcd for C₁₂H₂₇O₂Si (M+H)⁺: 231, Found: 231.



Glacial acetic acid (52.8 μL, 924.4 μmol, 10.2 equiv relative to catalyst) was added to a red-orange solution of (S,S)-(+)-N,N'-Bis(3,5-di-*tert*-butylsalicylidene)-1,2-cyclohexanediaminocobalt(II) (52.9 mg, 87.62 μmol, 0.2 mol%) in 1.1 mL toluene at 23 °C in a flask open to the air. The resulting solution was stirred for 30 min at 23 °C and was then concentrated in vacuo to provide a brown solid. Epoxide (**rac-3**) (11.39 g, 43.40 mmol, 1.0 equiv) was added to this catalyst, and the resulting brown-black solution stirred under argon at 0 °C for 10 min before adding water (430.2 μL, 23.87 mmol, 0.55 equiv) to the reaction solution. The resulting mixture was stirred for 5 min at 0 °C before removing the cooling bath. After 28

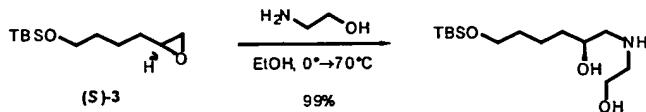
hr at 23 °C, the enantioenriched epoxide (*S*)-3 was distilled directly from the reaction mixture (95–99 °C, 0.5 mm Hg), affording epoxide (*S*)-3 as a clear oil (4.62 g, 46%) and the product diol as a red-brown oil (5.39 g, 50%, contaminated with catalyst and water). Both the epoxide and the diol were spectroscopically pure by ¹H NMR. Epoxide (*S*)-3 was determined to be 5 provided in >98% ee by Mosher ester analysis (¹⁹F NMR) of the 2° azidoalcohol provided by reaction of the epoxide with sodium azide.

(S)-3: ¹H NMR (400 MHz, CDCl₃), δ 3.62 (t, 2H, J = 6.0 Hz, CH₂OTBS), 2.91 (m, 1H, CH(O)CH₂), 2.75 (dd, 1H, J = 4.0, 5.2 Hz, CH(O)CH₂ (*cis*-)), 2.47 (dd, 1H, J = 2.8, 5.2 Hz, CH(O)CH₂ (*trans*-)), 1.61–1.47 (m, 6H, CH₂CH₂CH₂CH₂OTBS), 0.98 (s, 9H, SiC(CH₃)₃), 0.05 (s, 6H, Si(CH₃)₂). ¹³C NMR (100 MHz, CDCl₃), δ 63.1, 52.5, 47.3, 32.8, 32.5, 26.2, 22.6, 18.6, –5.0. FTIR (neat film), cm^{–1} 2951 (s), 2929 (s), 2856 (s), 1472 (s), 1255 (s), 1099 (s), 836 (s), 775 (s). R_f 0.73, 50% ethyl acetate–hexanes. LRMS (TOF-ES⁺) Calcd for C₁₂H₂₇O₂Si (M+H)⁺: 231, Found: 231.

15

Diol: ¹H NMR (400 MHz, CDCl₃), δ 3.72 (m, 1H, CH₂CH(OH)CH₂), 3.66 (ddd, 1H, J = 3.2, 6.4, 10.8 Hz, CH₂OH), 3.63 (t, 2H, J = 6.0 Hz, TBSOCH₂), 3.44 (ddd, 1H, J = 4.8, 7.6 Hz, 12.4 Hz, CH₂OH), 2.11 (m, 1H, CH₂CH(OH)), 1.86 (m, 1H, CH₂CH(OH)), 1.51–1.58 (m, 2H, TBSOCH₂CH₂), 1.45–1.49 (m, 2H, CH₂CH₂CH(OH)), 0.89 (s, 9H, SiC(CH₃)₃), 0.05 (s, 6H, Si(CH₃)₂). ¹³C NMR (100 MHz, CDCl₃), δ 72.4, 67.0, 63.2, 33.1, 32.8, 26.2, 22.1, 18.6, –5.0. FTIR (neat film), cm^{–1} 3372 (br, s, OH), 2935 (s, CH), 2857 (s, CH), 1472 (m), 1254 (m), 1102 (m), 836 (m), 775 (m). R_f 0.21, 50% ethyl acetate–hexanes. HRMS (TOF-ES⁺) Calcd for C₁₂H₂₉O₃Si (M+H)⁺: 249.1886, Found: 249.1877.

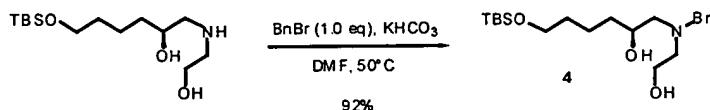
25



A solution of epoxide (*S*)-3 (4.78 g, 20.75 mmol, 1.0 equiv) in 207 mL abs. EtOH was stirred at 0 °C for 10 min. Ethanolamine (62.6 mL, 1.04 mol, 50.0 equiv), was then added

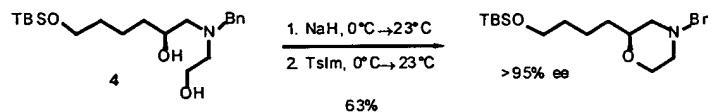
gradually to the clear reaction solution over 10 min, providing a yellow solution which was allowed to warm to 23 °C. This solution was at 70 °C for 1 hr. Ethanol was removed from the reaction solution in vacuo and the resulting yellow oil partitioned between 700 mL ethyl acetate and 700 mL water. The separated organic layer was then washed with 1×500 mL water and 5 1×500 mL brine, dried over potassium carbonate, and concentration in vacuo to provided analytically pure aminodiol as a yellowish transparent oil (5.99 g, 99%).

¹H NMR (400 MHz, CDCl₃), δ 3.68 (t, 2H, J = 5.2 Hz, CH₂OH), 3.65 (m, 1H, CH(OH)), 3.62 (t, 2H, J = 6.4 Hz, TBSOCH₂), 2.80 (q, 2H, J = 5.6 Hz, CH₂CH₂OH), 2.74 (dd, 1H, J = 3.2, 10 12.4 Hz, CH(OH)CH₂NH), 2.51 (dd, 1H, J = 9.2, 12.0 Hz, CH(OH)CH₂NH), 1.50-1.58 (m, 2H, CH₂CH(OH)), 1.37-1.49 (m, 4H, TBSOCH₂CH₂CH₂), 0.89 (s, 9H, SiC(CH₃)₃), 0.05 (s, 6H, Si(CH₃)₂). ¹³C NMR (100 MHz, CDCl₃), δ 70.1, 63.3, 61.5, 55.2, 51.2, 35.0, 33.0, 26.2, 22.2, 18.6, -5.0. FTIR (neat film), cm⁻¹ 3304 (br, s, OH), 2929 (s), 2857 (s), 1471 (m), 1234 (m), 1098 (m). R_f 0.35, 50% ethyl acetate–hexanes. HRMS (TOF-ES⁺) Calcd for C₁₄H₃₄NO₃Si 15 (M+H)⁺: 292.2308, Found: 292.2297.



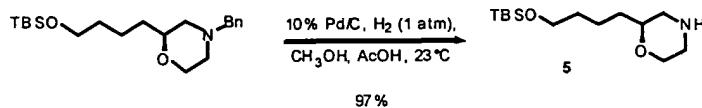
Potassium bicarbonate (6.93 g, 69.17 mmol, 2.0 equiv) was added to a solution of 20 substrate aminodiol (10.08 g 34.58 mmol, 1.0 equiv) in 300 mL N,N-dimethylformamide. Benzyl bromide (4.20 mL, 34.58 mmol, 1.0 equiv) was then added to the vigorously stirred resulting white suspension, the reaction vessel wrapped in foil, and the reaction vessel heated at 50 °C for 2.3 hr. After cooling, the reaction mixture was partitioned between 700 mL dichloromethane and 700 mL water and the organic layer was separated. The aqueous layer was 25 sequentially extracted with 1×500 mL, 1×300 mL, and 1×200 mL dichloromethane. All the organic extracts were then combined, washed with 1×1.6 L water, and dried over sodium sulfate. Concentration of the dried extracts in vacuo provided benzyl amine 4 as a viscous yellow oil which did not require further purification (12.60 g, 92%).

4: ^1H NMR (400 MHz, CDCl_3), 1.3:1 mixture of rotamers, * indicates minor rotamer, δ 7.27-7.35 (m, 5H, C_6H_5), 3.868 (s, 1H*, CH_2^*Ph), 3.83 (s, 1H, CH_2Ph), 3.64-3.71 (m, 2H, 2H*, CH_2OH , CH_2^*OH), 3.59 (t, 2H, $J = 6.4$ Hz, TBSOCH_2), 3.58 (s, 1H, CH_2Ph), 3.56 (s, 1H*, CH_2^*Ph), 2.82 (dd, 1H*, $\text{CH}_2^*\text{CH}_2\text{OH}$), 2.80 (dd, 1H, $\text{CH}_2\text{CH}_2\text{OH}$), 2.65 (t, 1H, $\text{CH}_2\text{CH}_2\text{OH}$),
5 2.61 (t, 1H*, $\text{CH}_2^*\text{CH}_2\text{OH}$), 2.56 (dd, 1H, $J = 3.2, 13.2$ Hz, $\text{CH}(\text{OH})\text{CH}_2\text{N}$), 2.47 (dd, 1H, $J = 10.0, 12.8$ Hz, $\text{CH}(\text{OH})\text{CH}_2\text{N}$), 1.47-1.54 (m, 4H, $\text{TBSOCH}_2\text{CH}_2\text{CH}_2\text{CH}_2$), 1.34-1.42 (m, 2H, $\text{TBSOCH}_2\text{CH}_2\text{CH}_2$), 0.89 (s, 9H, $\text{Si}(\text{CH}_3)_3$), 0.04 (s, 6H, $\text{Si}(\text{CH}_3)_2$). ^{13}C NMR (100 MHz, CDCl_3), δ 138.6, 129.0, 128.6, 127.4, 68.2, 63.3, 61.0, 60.1, 59.8, 56.4, 34.8, 33.1, 26.3, 22.2, 18.7, -4.9. FTIR (neat film), cm^{-1} 3362 (br, m, OH), 2924 (m), 2856 (m), 1460 (m), 1249 (m),
10 1092 (m), 834 (m). R_f 0.34, 70% ethyl acetate–hexanes. HRMS (TOF-ES $^+$) Calcd for $\text{C}_{21}\text{H}_{40}\text{NO}_3\text{Si} (\text{M}+\text{H})^+$: 382.2777, Found: 382.2760.



15 A solution of N-benzyl diol 4 (1.54 g, 4.02 mmol, 1.0 equiv) in 40.0 mL tetrahydrofuran was stirred at 0 °C for 10 min and was then added via cannula to sodium hydride (254.2 mg, 10.06 mmol, 2.5 equiv). The resulting white suspension was stirred vigorously at 0 °C for 5 min before removing the cooling bath. After 1 hr at 23 °C, the reaction suspension was returned to a 0 °C bath for 10 min and N-tosylimidazole (894.5 mg, 4.02 mmol, 1.0 equiv) added in 3 portions over 12 min (gas evolution was observed subsequent to the addition of each portion). After stirring the resulting reaction solution for a further 10 min at 0 °C, the cooling bath was again removed. After 1 hr at 23 °C, excess sodium hydride was CAREFULLY quenched by the slow addition of 30 mL saturated aqueous ammonium chloride to the reaction suspension at 0 °C. The reaction mixture was then partitioned between 340 mL ammonium chloride (sat., aq.) and 370 mL diethyl ether and the organic layer separated and washed further with 1×300 mL water and 1×200 mL brine. The aqueous washes were combined and extracted with 2×200 mL diethyl ether. All of the organic extracts were then combined and dried over sodium sulfate. Concentration of the extracts in vacuo provided a yellow oil, which was purified by flash column chromatography (SiO_2 , 20% ethyl acetate–hexanes), affording the N-benzyl morpholine product
20 as a yellow oil (926.6 mg, 63%).
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¹H NMR (400 MHz, CDCl₃), 1:1 mix of rotamers, * indicates rotamer, δ 7.29-7.34 (m, 5H, C₆H₅), 3.83 (ddd, 1H, J = 1.6, 2.8, 12.8 Hz, CHOCH₂), 3.64 (dt, 1H, J = 2.0, 11.2 Hz, CHOCH₂), 3.58 (t, 2H, J = 6.0 Hz, TBSOCH₂), 3.49 (s + m, 2H, CH₂Ph, CH(OR)CH), 3.48 (s, 1H*, CH₂*Ph), 2.72 (d, 1H, J = 11.6 Hz, CH(OR)CH₂N), 2.65 (dd, 1H, J = 1.6, 11.6 Hz, OCH₂CH₂N), 2.14 (dt, 1H, J = 3.2, 11.6 Hz, OCH₂CH₂N), 1.84 (t, 1H, J = 11.2 Hz, CH(OR)CH₂N), 1.41-1.54 (m, 4H, TBSOCH₂CH₂CH₂CH₂), 1.30-1.41 (m, 2H, TBSOCH₂CH₂CH₂), 0.88 (s, 9H, SiC(CH₃)₃), 0.03 (s, 6H, Si(CH₃)₂). ¹³C NMR (100 MHz, CDCl₃), δ 129.2, 128.3, 127.2, 75.8, 67.0, 63.5, 63.3, 58.9, 53.4, 33.7, 33.1, 26.2, 21.9, 18.7, -4.9. FTIR (neat film), cm⁻¹ 3372 (w), 2929 (s), 2856 (s), 1454 (m), 1255 (m), 1101 (s), 835 (s). R_f 0.68, 70% ethyl acetate-hexanes. LRMS (TOF-ES⁺) Calcd for C₂₁H₃₈NO₂Si (M+H)⁺: 364, Found: 364.

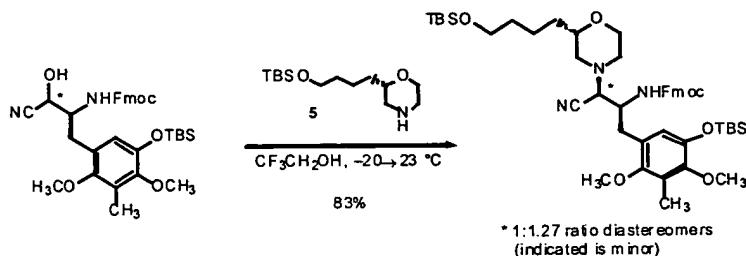


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Glatial acetic acid (379.0 μL, 6.63 mmol, 2.6 equiv) was added to a solution of N-benzyl morpholine substrate (926.6 mg, 2.55 mmol, 1.0 equiv) in 30.0 mL methanol. 10% Palladium on activated carbon (271.2 mg, 254.8 μmol (Pd), 0.1 equiv), was then added to the clear solution. The resulting black suspension was cycled under a H₂ atmosphere (1 atm) by alternately evacuating the reaction vessel and refilling with H_{2(g)} (5×). After stirring 2 hr under 1 atm hydrogen, the reaction suspension was filtered through Celite 545 (CAUTION: do not allow the catalyst to become dry – ignition hazard), the catalyst washed with 3×10 mL methanol, and the filtrate partitioned between 100 mL diethyl ether and 100 mL sodium bicarbonate (sat., aq.). The organic layer was then washed with a further 1×80 mL brine and the combined aqueous washes extracted with 1×100 mL diethyl ether. All organic extracts were then combined, dried over potassium carbonate, and concentrated in vacuo to provide spectroscopically pure siloxymorpholine 5 as an iridescent yellow-tinged oil (679.0 mg, 97%). Mosher amide analysis of siloxymorpholine 5 (¹H NMR) indicated that the product was provided in >95% ee.

5: ^1H NMR (400 MHz, CDCl_3), δ 3.87 (dt, 1H, $J = 2.0, 11.6$ Hz, CHOCH_2), 3.65 (ddd, 1H, $J = 7.2, 12.0, 14.0$ Hz, CHOCH_2), 3.60 (t, 2H, $J = 6.4$ Hz, TBSOCH_2), 3.47 (m, 1H, $\text{CH}(\text{OR})$), 2.94 (dd, 1H, $J = 2.4, 12.4$ Hz, $\text{CH}(\text{OR})\text{CH}_2$), 2.88 (dd, 2H, $J = 2.8, 8.0$ Hz, $\text{OCH}_2\text{CH}_2\text{N}$), 2.56 (dd, 1H, $J = 10.4, 12.4$ Hz, $\text{CH}(\text{OR})\text{CH}_2\text{N}$), 1.36-1.54 (m, 6H, 5 $\text{TBSOCH}_2\text{CH}_2\text{CH}_2\text{CH}_2$), 0.88 (s, 9H, $\text{SiC}(\text{CH}_3)_3$), 0.04 (s, 6H, $\text{Si}(\text{CH}_3)_2$). ^{13}C NMR (100 MHz, CDCl_3), δ 68.3, 63.2, 51.6, 46.2, 33.7, 33.0, 26.1, 21.7, 18.5, -5.1. FTIR (neat film), cm^{-1} 3342 (w, NH), 2932 (s), 2856 (s), 1472 (m), 1461 (m), 1255 (m), 1094 (s), 835 (s). R_f 0.43, 10% methanol-dichloromethane, triethylamine-dipped plate. HRMS (TOF-ES $^+$) Calcd for $\text{C}_{14}\text{H}_{32}\text{NO}_2\text{Si} (\text{M}+\text{H})^+$: 274.2202, Found: 274.2191.

10



To a solution of siloxymorpholine 5 (606.6 mg, 2.22 mmol, 5.0 equiv) in 4.5 mL 2,2,2-trifluoroethanol was stirred over a -20 °C bath for 10 min and was then added by cannula to substrate cyanohydrin (Prepared as previously reported: Myers, A. G.; Kung, D. W.; Zhong, B.; Movassaghi, M.; and Kwon, S. *J. Am. Chem. Soc.* **1999**, *121*, 8401–8402; Myers, A. G.; Kung, D. W. *J. Am. Chem. Soc.* **1999**, *121*, 10828–10829) (273.4 mg, 453.56 μmol , 1.0 equiv) at -20 °C. The resulting yellow solution was stirred for 5 min at -20 °C, then allowed to warm to 23 °C. After 3 hr at 23 °C, the reaction solution was concentrated in vacuo, then from 1×5.0 mL 15 benzene. Purification of the concentrate by flash column chromatography (SiO_2 , 18% ethyl acetate–hexanes) afforded the *syn*-mopholinonitrile as a clear oil (149.6 mg, 38%) and the *anti*-mopholinonitrile as a clear oil (175.0 mg, 45%). Siloxymorpholine 5 was recovered from the 20 column by elution with 10% methanol–dichloromethane + 2% (v/v) triethylamine, affording recovered siloxymorpholine 5 as a yellow oil (493.9 mg, 100%).

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***syn*-mopholinonitrile** (minor): ^1H NMR (400 MHz, CDCl_3), δ 7.76 (d, 2H, $J = 7.6$ Hz, ArH), 7.54 (d, 2H, $J = 7.2$ Hz, ArH), 7.40 (t, 2H, $J = 7.2$ Hz, ArH), 7.30 (t, 2H, $J = 7.2$ Hz, ArH),

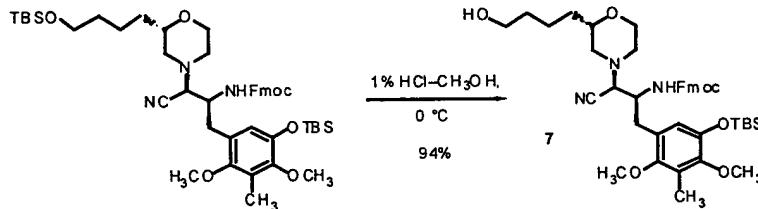
6.54 (s, 1H, ArH), 5.29 (d, 1H, J = 6.8 Hz, NHFmoc), 4.38 (app. t, 1H, J = 7.6 Hz, CO₂CH₂),
4.27 (app. t, 1H, J = 10.8 Hz, CO₂CH₂), 4.20 (t, 1H, J = 6.8 Hz, CO₂CH₂CH), 4.18 (br s, 1H,
OCHCH₂), 3.86 (d, 1H, J = 11.6 Hz, OCH₂CH₂), 3.74 (s, 3H, ArOCH₃), 3.71 (s, 3H, ArOCH₃),
3.57 (t, 2H, TBSOCH₂), 3.50–3.40 (m, 2H, OCH₂CH₂, CHCN), 3.37 (m, 1H, CHNHFmoc),
5 3.10 (d, 1H, J = 11.2 Hz, NCH₂), 2.94 (m, 1H, NCH₂), 2.86 (d, 1H, J = 11.2 Hz, NCH₂), 2.52 (d,
1H, 9.6 Hz, CH₂Ar), 2.30 (t, 1H, J = 10.4 Hz, CH₂Ar), 2.22 (s, 3H, ArCH₃), 1.52–1.26 (m, 6H,
OCH₂CH₂CH₂CH₂), 0.99 (s, 9H, ArOSiC(CH₃)₃), 0.88 (s, 9H, ROSiC(CH₃)₃), 0.16, 0.15 (2s,
6H, ArOSi(CH₃)₂), 0.03 (s, 6H, ROSi(CH₃)₂). ¹³C NMR (100 MHz, CDCl₃), δ 151.5, 149.7,
145.4, 144.0, 141.5, 128.0, 127.3, 126.2, 125.4, 125.2, 123.7, 121.0, 120.3, 115.0, 75.8, 67.0,
10 66.7, 63.2, 61.9, 60.8, 60.1, 58.1, 50.9, 48.2, 47.3, 33.4, 33.0, 31.8, 26.2, 25.9, 21.7, 18.6, 18.4,
10.2, –4.3, –5.0. FTIR (neat film), cm^{–1} 3352 (w, NH), 2931 (s), 2856 (s), 1727 (s, NCO₂), 1482
(m), 1255 (s), 1238 (s), 1103 (m), 1066 (s), 838 (s). R_f 0.53, 30% ethyl acetate–hexanes. HRMS
(TOF-ES⁺) Calcd for C₄₈H₇₂N₃O₇Si₂ (M+H)⁺: 858.4909, Found: 858.4890.

15 epi-syn-morpholinonitrile (trace): ¹H NMR (400 MHz, CDCl₃), δ 7.75 (d, 2H, J = 7.6
Hz, ArH), 7.57 (dd, 2H, J = 2.8, 7.2 Hz, ArH), 7.39 (t, 2H, J = 7.6 Hz, ArH), 7.30 (t, 2H, J = 7.2
Hz, ArH), 6.49 (s, 1H, ArH), 5.39 (d, 1H, J = 8.4 Hz, NHFmoc), 4.34–4.31 (m, 2H, CO₂CH₂),
4.20 (t, 1H, J = 7.6 Hz, CO₂CH₂CH), 4.13 (m, 1H, OCHCH₂), 3.97 (d, 1H, J = 10.8 Hz,
ROCH₂CH₂), 3.72 (s, 3H, ArOCH₃), 3.67 (s, 4H, ArOCH₃, CHCN), 3.61 (t, 3H, J = 6.4 Hz,
20 TBSOCH₂, ROCH₂CH₂), 3.53 (m, 1H, CHNHFmoc), 3.03 (dd, 1H, J = 4.0, 14.0 Hz, NCH₂),
2.87 (dd, 1H, J = 7.6, 13.6 Hz, NCH₂), 2.81 (d, 1H, J = 11.6 Hz, NCH₂), 2.57 (d, 1H, J = 10.0
Hz, CH₂Ar), 2.52 (dd, 1H, J = 2.8, 11.2 Hz, NCH₂), 2.34 (t, 1H, J = 10.4 Hz, CH₂Ar), 2.21 (s,
3H, ArCH₃), 1.58–1.34 (m, 6H, TBSOCH₂CH₂CH₂CH₂), 0.97 (s, 9H, ArOSiC(CH₃)₃), 0.89 (s,
9H, ROSiC(CH₃)₃), 0.12 (app. d, 6H, J = 6.8 Hz, ArOSi(CH₃)₂), 0.05 (s, 6H, ROSi(CH₃)₂).
25 FTIR (neat film), cm^{–1} 3346 (w, NH), 2932 (s), 2856 (s), 1721 (s, NCO₂), 1481 (m), 1451 (m),
1253 (s), 1104 (m), 1066 (s), 1010 (m), 838 (s). R_f 0.49, 30% ethyl acetate–hexanes. HRMS
(TOF-ES⁺) Calcd for C₄₈H₇₂N₃O₇Si₂ (M+H)⁺: 858.4909, Found: 858.4890.

30 anti-morpholinonitrile (major): ¹H NMR (400 MHz, CDCl₃), δ 7.76 (d, 2H, J = 7.2 Hz,
ArH), 7.60–7.52 (m, 2H, ArH), 7.40 (t, 2H, J = 7.6 Hz, ArH), 7.30 (dt, 2H, J = 1.2, 7.6 Hz,
ArH), 6.50 (s, 2H, ArH), 5.44 (d, 1H, J = 8.0 Hz, NHFmoc), 4.33 (d, 2H, J = 7.6 Hz, CO₂CH₂),

4.21 (t, 1H, J = 7.2 Hz, $\text{CO}_2\text{CH}_2\text{CH}$), 4.17 (m, 1H, OCHCH_2), 3.90 (d, 1H, J = 10.0 Hz, OCH_2CH_2), 3.72 (s, 3H, ArOCH_2), 3.67 (s, 3H, ArOCH_3), 3.63 (d, 1H, CHCN), 3.61 (t, 2H, J = 6.4 Hz, TBSOCH_2), 3.55 (t, 1H, J = 6.4 Hz, OCH_2CH_2), 3.51 (s, 1H, CHNFmoc), 3.02 (dd, 1H, J = 4.0, 13.6 Hz, NCH_2), 2.89 (dd, 1H, J = 7.6, 14.0 Hz, NCH_2), 2.85 (d, 1H, J = 10.0 Hz, 5 NCH_2), 2.64 (dt, 1H, J = 11.6 Hz, NCH_2), 2.52 (d, 1H, J = 10.4 Hz, CH_2Ar), 2.25 (t, 1H, J = 10.8 Hz, CH_2Ar), 2.21 (s, 3H, ArCH_3), 1.56–1.36 (m, 6H, $\text{OCH}_2\text{CH}_2\text{CH}_2\text{CH}_2$), 0.98–0.96 (m, 9H, 10 $\text{ArOSi}(\text{CH}_3)_3$), 0.89–0.86 (m, 9H, $\text{ROSi}(\text{CH}_3)_3$), 0.15–0.12 (m, 6H, $\text{ArOSi}(\text{CH}_3)_2$), 0.05–0.02 (m, 6H, $\text{ROSi}(\text{CH}_3)_2$). ^{13}C NMR (100 MHz, CDCl_3), δ 156.1, 151.4, 149.6, 145.3, 144.1, 141.4, 127.8, 127.2, 126.1, 125.3, 124.3, 120.9, 120.8, 120.1, 75.8, 71.3, 67.4, 66.7, 63.2, 60.6, 60.0, 53.1, 47.2, 33.6, 33.4, 33.0, 32.9, 31.7, 29.9, 26.1, 25.8(2), 21.8, 18.3, 10.1, –4.4(2), –5.1. FTIR (neat film), cm^{-1} 3341 (w, NH), 2929 (s), 2857 (m), 1725 (s), 1481 (s), 1253 (s), 1237 (s), 1065 (s), 838 (s). R_f 0.41, 30% ethyl acetate–hexanes. HRMS (TOF-ES $^+$) Calcd for $\text{C}_{48}\text{H}_{72}\text{N}_3\text{O}_7\text{Si}_2$ ($\text{M}+\text{H}$) $^+$: 858.4909, Found: 858.4873.

15

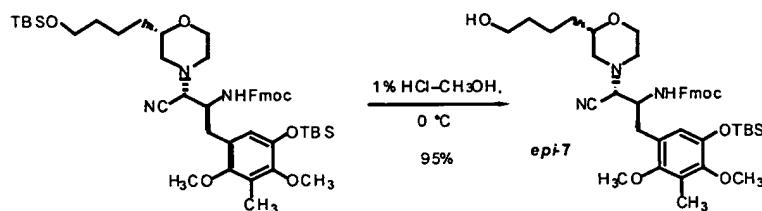


Concentrated hydrochloric acid (240.0 μL , 2.88 mmol, 1% v/v with methanol, ~6 equiv) was added to a solution of *syn*-aminonitrile substrate (406.2 mg, 473.3 μmol , 1.0 equiv) in 24.0 mL methanol that had been stirred at 0 °C for 10 min. After 5 min, the reaction solution was 20 partitioned between 240 mL diethyl ether and 240 mL (3:1) water : sodium bicarbonate (sat, aq). The organic layer was isolated and washed with 1×150 mL brine. The combined aqueous layers were then further extracted with 1×250 mL diethyl ether and the combined organic extracts dried over sodium sulfate. Concentration in vacuo followed by column chromatography (SiO_2 , 60% ethyl acetate–hexanes) provided alcohol 7 as a clear oil (330.5 mg, 94%).

25

7: ^1H NMR (400 MHz, CDCl_3), δ 7.76 (d, 2H, J = 8.0 Hz, ArH), 7.54 (d, 2H, J = 7.6 Hz, ArH), 7.40 (t, 2H, J = 8.0 Hz, ArH), 7.30 (t, 2H, J = 7.6 Hz, ArH), 6.55 (s, 1H, ArH), 5.30 (app d, 1H, NHFmoc), 4.39 (m, 1H, OCH_2CH), 4.27 (m, 1H, OCH_2CH), 4.20 (t, 1H, J = 6.4 Hz,

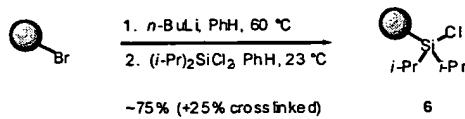
OCH₂CH), 3.87 (d, 1H, J = 10.8 Hz, CH₂CHO), 3.74 (s, 3H, ArOCH₃), 3.72 (s, 3H, ArOCH₃), 3.66 (d, 1H, J = 6.8 Hz, CHCN), 3.61 (t, 2H, J = 5.6 Hz, HOCH₂), 3.50 (d, 1H, J = 11.2 Hz, ROCH₂CH₂), 3.44 (d, 1H, J = 9.2 Hz, ROCH₂CH₂), 3.37 (m, 1H, CHNHFmoc), 3.09 (d, 1H, J = 13.2 Hz, CH₂Ar), 2.93 (m, 1H, CH₂NCH₂), 2.87 (m, 1H, CH₂NCH₂), 2.52 (m, 1H, CH₂NCH₂), 5 2.42 (m, 1H, CH₂NCH₂), 2.32 (m, 1H, CH₂Ar), 2.22 (s, 3H, ArCH₃), 1.54-1.33 (m, 6H, HOCH₂CH₂CH₂CH₂), 0.99 (s, 9H, C(CH₃)₃), 0.15, 0.16 (app d, 6H, Si(CH₃)₂). ¹³C NMR (100 MHz, CDCl₃), δ 175.2, 156.2, 151.5, 149.7, 144.0, 141.5, 128.0, 127.2, 126.2, 125.2, 123.7, 120.9, 120.3, 115.9, 75.7, 67.1, 66.7, 62.9, 61.9, 60.8, 60.1, 58.0, 50.9, 48.2, 47.3, 33.2, 32.8, 31.8, 25.9, 21.7, 184, 10.2, -4.3. FTIR (neat film), cm⁻¹ 3348 (br s, OH), 2934 (s), 2858 (s), 10 1721 (s, NHCO₂), 1482 (m), 1238 (m), 1064 (m), 869 (m), 738 (m). R_f 0.50, 70% ethyl acetate–hexanes. LRMS (TOF-ES⁺) Calcd for C₃₆H₄₃N₃NaO₇ (M-TBS+Na)⁺: 651, Found: 651.



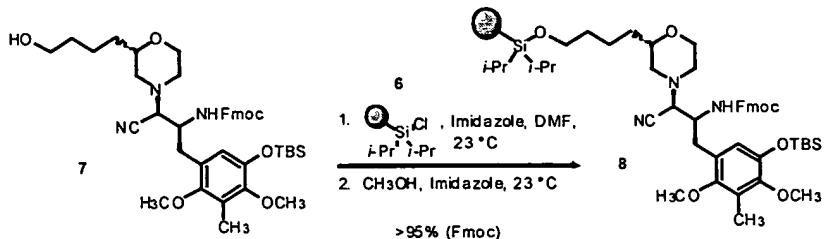
15 Concentrated hydrochloric acid (250.0 μL, 3.00 mmol, 1% v/v with methanol, ~3 equiv) was added to a solution of *anti*-aminonitrile substrate (916.2 mg, 1.07 mmol, 1.0 equiv) in 25.0 mL methanol that had been stirred at 0 °C for 10 min. After 5 min, the reaction solution was partitioned between 250 mL diethyl ether and 250 mL (3:1) water : sodium bicarbonate (sat, aq). The organic layer was isolated and washed with 1×150 mL brine. The combined aqueous layers 20 were then further extracted with 2×250 mL diethyl ether and the combined organic extracts dried over sodium sulfate. Concentration in vacuo followed by column chromatography (SiO₂, 60% ethyl acetate–hexanes) provided alcohol *epi*-7 as a clear oil (756.5 mg, 95%).

epi-7: ¹H NMR (400 MHz, CDCl₃), δ 7.76 (d, 2H, J = 7.2 Hz, ArH), 7.58 (dd, 2H, J = 4.0, 7.2 Hz, ArH), 7.38 (t, 2H, J = 7.6 Hz, ArH), 7.31 (dt, 2H, J = 0.8, 7.2 Hz, ArH), 6.50 (s, 1H, ArH), 5.47 (d, 1H, J = 8.8 Hz, NHFmoc), 4.33 (d, 2H, J = 7.2 Hz, CO₂CH₂), 4.21 (t, 1H, J = 7.2 Hz, CO₂CH₂CH), 4.18 (m, 1H, ROCH₂), 3.91 (d, 1H, J = 9.6 Hz, OCHCH₂), 3.72 (s, 3H, ArOCH₃), 3.67 (s, 3H, ArOCH₃), 3.66–3.61 (m, 4H, HOCH₂, ROCH₃, CHCN), 3.51 (m, 1H,

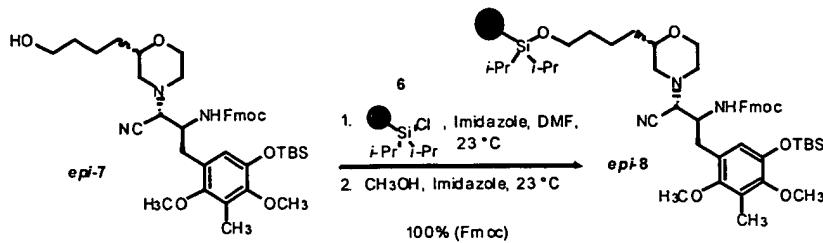
CHNHFmoc), 3.03 (dd, 1H, J = 4.0, 14.0 Hz, NCH2), 2.92–2.84 (m, 2H, NCH2, CH2Ar), 2.64 (dt, 1H, J = 2.8, 10.8 Hz, NCH2), 2.52 (d, 1H, J = 10.8 Hz, NCH2), 2.27 (d, 1H, J = 10.4 Hz, CH2Ar), 2.21 (s, 3H, ArCH3), 1.61–1.36 (m, 6H, HOCH2CH2CH2CH2), 0.97 (s, 9H, SiC(CH3)3), 0.13 (app. d, 6H, J = 4.4 Hz, Si(CH3)2). ^{13}C NMR (100 MHz, CDCl3), δ 156.2, 5 151.5, 149.7, 145.4, 144.2, 144.0, 141.5, 127.9, 127.3, 126.2, 125.5, 125.4, 124.4, 121.0, 120.2, 115.6, 75.9, 67.5, 66.8, 62.9, 61.9, 60.7, 60.1, 53.0, 52.7, 51.3, 47.3, 33.5, 32.8, 32.1, 25.9, 21.8, 18.4, 10.2, –4.3. FTIR (neat film), cm^{-1} 3335 (m, NH/OH), 2933 (s), 2858 (m), 1714 (s, NCO2), 1481 (s), 1451 (m), 1236 (s), 1117 (s), 1064 (m), 1012 (s), 867 (s), 739 (m). R_f 0.46, 70% ethyl acetate–hexanes. HRMS (TOF-ES $^+$) Calcd for C42H58N3O7Si ($M+H$) $^+$: 744.4044, Found: 10 744.4060.



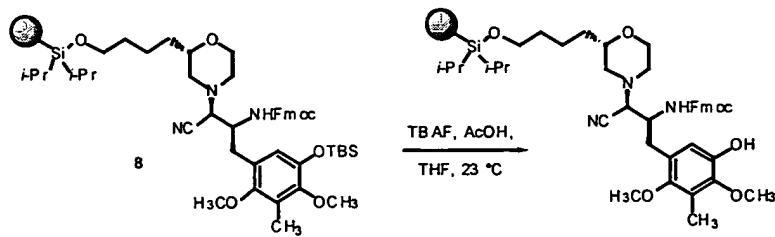
Polystyrene chlorodiisopropylsilane **6** was prepared by a modification of the procedure reported by Danishefsky (Randolph, J. T.; McClure, K. F.; Danishefsky, S. J. *J. Am. Chem. Soc.* 1995, 117, 5712–5719). Commercial 4-bromopolystyrene (Novabiochem, 4-bromo polystyrene HL (P/N: 01-64-0255), 50–100 mesh, Calbiochem-Novabiochem Corp., 10394 Pacific Center Court, San Diego, CA 92121) (3.65 g, 7.12 mmol, 1.95 mmol/g, 1.0 equiv) was washed with 2×35 mL benzene under an argon atmosphere in a modified round-bottomed flask bearing an integral glass vacuum frit and was then resuspended in 35 mL benzene. *n*-Butyl lithium (2.50 M in hexanes, 7.72 mL, 19.29 mmol, 2.71 equiv) was added to this suspension and the resulting mixture stirred at 60 °C for 3 hr. The cloudy white reaction supernatant was then removed by vacuum filtration and the resin washed with 1×35 mL benzene. The resin was then resuspended in 35 mL benzene and dichlorodiisopropylsilane (5.14 mL, 28.47 mmol, 4.0 equiv) added. The resulting suspension was stirred for 3 hr at 23 °C and the reaction supernatant again removed via filtration. The resin was then sequentially washed with 2×36 mL (5:1) benzene:acetonitrile, 1×35 mL benzene, 3×35 mL *N,N*-dimethylformamide, 2×35 mL tetrahydrofuran, and 2×35 mL dichloromethane and dried in vacuo for 8 hr to provide polystyrene chlorodiisopropylsilane **6** as a yellowish-white, free-flowing resin (3.98 g).



A solution of alcohol 7 (54.1 mg, 72.7 μmol , 1.0 equiv) in 3.6 mL N,N-dimethylformamide was added via cannula to a mixture of polystyrene chlorodiisopropylsilane 6 (412.3 mg, 566.1 μmol , 7.8 equiv, 1.373 mmol/g) and imidazole (44.6 mg, 654.4 μmol , 9.0 equiv) at 23 °C. After 2 hrs, additional imidazole (89.1 mg, 1.31 mmol, 18.0 equiv) and methanol (117.8 μL , 2.91 μmol , 40.0 equiv) were added to the resulting clear suspension of yellowish resin and the resulting mixture stirred for 15 hr. The reaction supernatant was then removed by crimped cannula, and the product resin washed sequentially with 3×4 mL N,N-dimethylformamide, 3×4 mL tetrahydrofuran, and 2×4 mL dichloromethane, removing the wash solution via crimped cannula. Drying of the resin in vacuo for 5 hr provided a free-flowing yellowish-white resin. The yield of this transformation was determined by cleavage of the 9-fluorenylmethoxycarbonyl (Fmoc) protecting group (20% piperidine–N,N-dimethylformamide, 2 min) from a measured aliquot of resin followed by UV quantitation of the liberated dibenzofulvene chromophore (A_{290}) according to the established procedure (See, for example, 1999 Novabiochem Catalog & Peptide Synthesis Handbook, Calbiochem–Novabiochem Corporation, 10394 Pacific Center Court, San Diego, CA 92121, pp. S43). Averaging two replicate measurements gave a resin loading level of 0.1502 mmol/g (95%). (Note: trace Fmoc cleavage during the loading and capping reactions suggests >95% loading.) Chromatographic separation of material cleaved from the product resin by incubating ~5 mg samples of washed resin in a mixture of 100 μL CH_2Cl_2 , 20 μL CH_3OH , and 10 μL conc. hydrochloric acid for 10 min indicated the solely the presence of substrate alcohol 7, confirming the clean formation of product resin 8.



A solution of alcohol *epi*-7 (756.5 mg, 1.02 mmol, 1.0 equiv) in 40.0 mL N,N-dimethylformamide was added via cannula (along with 2×5.0 mL N,N-dimethylformamide washes) to a mixture of polystyrene chlorodisopropylsilane **6** (3.98 g, 5.34 mmol, 5.24 equiv, 1.34 mmol/g) and imidazole (432.6 mg, 6.35 mmol, 6.25 equiv) at 23 °C. After 5 hrs, additional imidazole (1.25 g, 18.3 mmol, 18.0 equiv) and methanol (1.65 mL, 40.7 mmol, 40.0 equiv) were added to the resulting clear suspension of yellowish resin and the resulting mixture stirred for 16 hr. The reaction supernatant was then removed by crimped cannula, and the product resin 10 washed sequentially with 3×40 mL N,N-dimethylformamide, 3×40 mL tetrahydrofuran, and 2×40 mL dichloromethane, removing the wash solution via crimped cannula. Drying of the resin in vacuo for 5 hr provided a free-flowing yellowish-white resin. The yield of this transformation was determined by cleavage of the 9-fluorenylmethoxycarbonyl (Fmoc) protecting group (20% piperidine–N,N-dimethylformamide, 2 min) from a measured aliquot of resin followed by UV 15 quantitation of the liberated dibenzofulvene chromophore (A_{290}) according to the established procedure.⁷ Averaging two replicate measurements gave a resin loading level of 0.2345 mmol/g (100%). Chromatographic separation of material cleaved from the product resin by incubating ~5 mg samples of washed resin in a mixture of 100 μ L CH₂Cl₂, 20 μ L CH₃OH, and 10 μ L conc. hydrochloric acid for 10 min indicated the solely the presence of substrate alcohol *epi*-7, 20 confirming the clean formation of product resin *epi*-8.

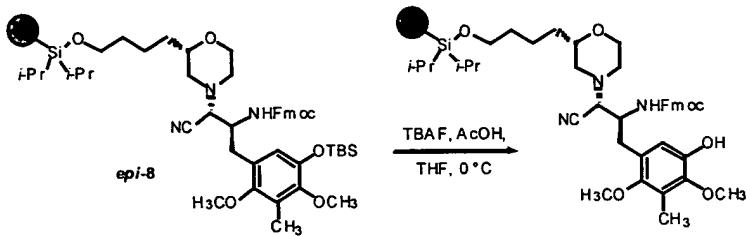


Acetic acid (253.8 μ L, 4.44 mmol, 10.0 equiv) was added to a suspension of aminonitrile resin **8** (2.12 g, 444.2 μ mol, 1.0 equiv, 0.1575 mmol/g) in 20.0 mL tetrahydrofuran. Tetra-*n*-butylammonium fluoride (1.0 M in tetrahydrofuran, 2.22 mL, 2.22 mmol, 5.0 equiv) was then added to the reaction suspension and the resulting suspension stirred at 23 °C for 1.5 hr. The 5 reaction supernatant was then removed via crimped cannula and the product resin washed sequentially with 3×20 mL N,N-dimethylformamide, 3×20 mL tetrahydrofuran, and 2×20 mL dichloromethane. The washed product resin was dried in vacuo overnight, affording a free-flowing yellow-orange resin. Photometric determination of the resin 9-fluorenylmethoxycarbonyl (Fmoc) loading level (A_{290} , 20% piperidine–N,N-dimethylformamide) gave a resin loading level of 0.09504 mmol/g, implying 45% Fmoc cleavage based on 100% yield for resin loading step. (TLC of the reaction supernatant confirms the formation of dibenzofulvene, indicating that Fmoc deprotection does occur under the reaction conditions.) Substrate was liberated from the product resin for solution-phase characterization by methanolysis of product resin aliquots (as described for **8** and *epi*-**8** above), isolation of the 10 supernatant solution (cannulation) followed by resin washing, aqueous workup of this substrate solution (sodium bicarbonate wash to neutralized hydrochloric acid), and chromatographic purification (90% ethyl acetate–hexanes).

15

¹H NMR (400 MHz, CDCl₃), δ 7.76 (d, 2H, J = 7.6 Hz, ArH), 7.54 (d, 2H, J = 7.2 Hz, 20 ArH), 7.40 (t, 2H, J = 7.6 Hz, ArH), 7.33 (t, 2H, J = 6.0 Hz, ArH), 6.64 (s, 1H, ArH), 5.48 (s, 1H, ArOH), 5.32 (app d, 1H, J = 6.4 Hz, NHFmoc), 4.32 (d, 2H, J = 6.8 Hz, CO₂CH₂), 4.21 (t, 1H, J = 6.8 Hz, CO₂CH₂CH), 4.16 (br s, 1H, CHOCH₂), 3.87 (d, 1H, J = 10.8 Hz, CHOCH₂), 3.77 (s, 3H, ArOCH₃), 3.73 (s, 3H, ArOCH₃), 3.63 (m, 2H, HOCH₂), 3.53–3.44 (m, 2H, CHCN, 25 CHOCH₂), 3.39 (m, 1H, CHNHFmoc), 3.06 (d, 1H, J = 13.6 Hz, OCH₂CH₂N), 2.96 (m, 1H, OCHCH₂N), 2.87 (d, 1H, J = 10.0 Hz, OCH₂CH₂N), 2.55 (t, 1H, J = 11.2 Hz, CH₂Ar), 2.43 (t, 1H, J = 10.4 Hz, OCH₂CH₂N), 2.30 (t, 1H, J = 8.4 Hz, CH₂Ar), 2.26 (s, 3H, ArCH₃), 1.60–1.30 (m, 6H, HOCH₂CH₂CH₂CH₂). ¹³C NMR (100 MHz, CDCl₃), δ 156.2, 150.6, 145.6, 145.4, 144.1, 143.9, 141.5, 128.0, 127.3, 125.3, 125.1, 120.3, 116.0, 115.3, 75.7, 67.1, 66.6, 63.0, 61.8, 61.0, 60.9, 57.8, 51.1, 48.7, 47.3, 33.2, 32.7, 31.6, 21.7, 10.3. FTIR (neat film), cm^{−1} 3347 (m, 30 NH), 2939 (s), 2864 (m), 1712 (s, NCO₂), 1483 (s), 1450 (s), 1233 (s), 1111 (s), 1051 (s), 1008

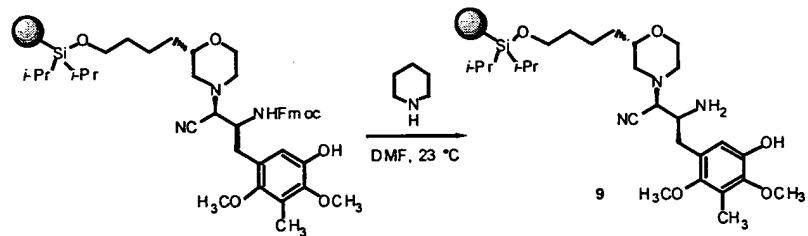
(m), 738 (s). R_f 0.14, 70% ethyl acetate–hexanes. HRMS (TOF-ES $^+$) Calcd for C₃₆H₄₄N₃O₇ (M+H) $^+$: 630.3179, Found: 630.3192.



5

Acetic acid (9.0 μ L, 156.8 μ mol, 2.2 equiv) and tetra-*n*-butylammonium fluoride (1.0 M in tetrahydrofuran, 78.4 μ L, 78.4 μ mol, 1.1 equiv) were sequentially added to a suspension of siloxane resin *epi*-8 (328.0 mg, 71.27 μ mol, 1.0 equiv) in 3.5 mL tetrahydrofuran that had been stirring at 0 °C for 10 min. After 4.5 hr, the reaction supernatant was removed via crimped cannula and the product resin washed with 2×4 mL N,N-dimethylformamide, 2×4 mL tetrahydrofuran, and 2×4 mL dichloromethane. The product resin was then dried in vacuo for 5 hr to provide a free-flowing, yellow resin. (Note: TLC of the reaction supernatant reveals the formation of dibenzofulvene, indicating that Fmoc deprotection occurs under the reaction conditions (see corresponding transformation of **8** above).)

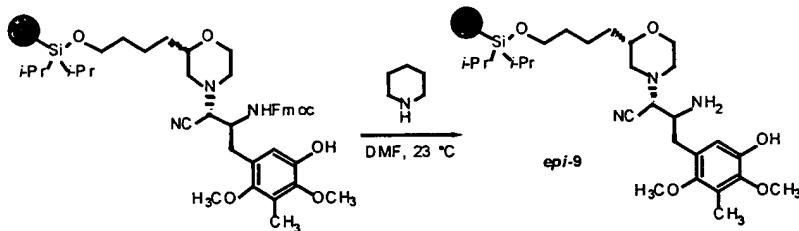
15



N-Fmoc-morpholinonitrile substrate resin (2.06 g, 443.5 µmol, 1.0 equiv, 0.2152 mmol/g) was suspended in 20.0 mL 20% (v/v) piperidine-N,N-dimethylformamide. The resulting suspension was stirred at 23 °C for 1.5 hr and the reaction supernatant then removed via crimped cannula. The product resin was then sequentially washed with 3×20 mL N,N-dimethylformamide, 3×20 mL tetrahydrofuran, and 2×20 mL dichloromethane and dried in

vacuo to provide product resin **9** as a free-flowing tan solid. Methanolysis of aliquots of washed product resin indicated the sole presence of the expected siloxane cleavage product.

9: ^1H NMR (400 MHz, CDCl_3), δ 6.68 (s, 1H, ArH), 3.93 (ddd, 1H, J = 1.6, 3.6, 11.2 Hz, 5 CHOCH_2), 3.78 (s, 3H, ArOCH₃), 3.70 (s, 3H, ArOCH₃), 3.64 (t, 2H, J = 6.4 Hz, HOCH₂), 3.59 (dt, 1H, J = 2.8, 11.6 Hz, CHOCH₂), 3.51 (m, 1H, CHNH₂), 3.38 (app dd, 1H, J = 2.8, 8.4 Hz, CHOCH₂), 3.24 (d, 1H, J = 10.0 Hz, CHCN), 3.07 (dd, 1H, J = 3.2, 13.2 Hz, OCHCH₂N), 2.64 (d, 1H, J = 11.2 Hz, OCH₂CH₂N), 2.56 (t, 1H, J = 12.4 Hz, CH₂Ar), 2.53 (dd, 1H, J = 6.4, 11.6 Hz, OCH₂CH₂N), 2.48 (dt, 1H, J = 3.6, 11.6 Hz, OCH₂CH₂N), 2.41 (t, 1H, J = 10.8 Hz, CH₂Ar), 10 2.24 (s, 1H, ArCH₃), 1.62–1.35 (m, 6H, HOCH₂CH₂CH₂CH₂). ^{13}C NMR (100 MHz, CDCl_3), δ 150.7, 145.5, 145.2, 126.3, 125.2, 115.8, 114.8, 75.9, 66.5, 64.5, 62.9, 60.9 (2), 58.3, 50.3, 47.2, 34.9, 33.2, 32.8, 21.7, 10.2. FTIR (neat film), cm^{-1} 3356 (br, m, NH, OH), 2940 (s, CH), 2862 (m), 1483 (m), 1458 (m), 1420 (m), 1113 (s), 1051 (m), 1010 (m), 911 (m), 732 (s). R_f 0.28, 15 10% methanol–dichloromethane, triethylamine-dipped plate. HRMS (TOF-ES⁺) Calcd for $\text{C}_{21}\text{H}_{34}\text{N}_3\text{O}_5$ ($\text{M}+\text{H}$)⁺: 408.2498, Found: 408.2516.

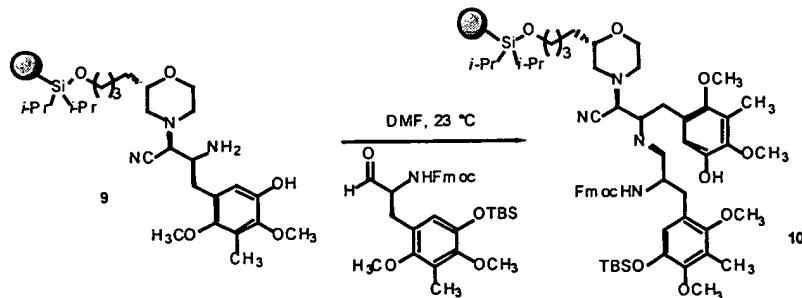


N-Fmoc-morpholinonitrile substrate resin (315.0 mg, 70.19 μmol , 1.0 equiv, 0.2228 mmol/g) was suspended in 3.95 mL 20% (v/v) piperidine-N,N-dimethylformamide. The resulting suspension was stirred at 23 °C for 5 hr and the reaction supernatant then removed via crimped cannula. The product resin was then sequentially washed with 3×4 mL N,N-dimethylformamide, 3×4 mL tetrahydrofuran, and 2×4 mL dichloromethane and dried in vacuo to provide product resin *epi*-9 as a free-flowing cream-colored resin. Solid-immobilized 20 substrate was liberated for characterization by suspending a sample of the product resin (179.0 mg, 41.96 μmol , 1.0 equiv) in a mixture of dichloromethane (5.0 mL), methanol (170.0 μL , 4.2 mmol, 100.0 equiv), and concentrated hydrochloric acid (17.1 μL , 205.6 μmol , 4.9 equiv) at 23 25 °C for 1 hr. The reaction mixture was then quenched with 10 mL water and extracted with dichloromethane (3×5 mL). The combined organic layers were dried over sodium sulfate and concentrated in vacuo to provide product resin *epi*-9 as a tan solid.

^oC over 22 hr. The reaction supernatant from this mixture was collected via crimped cannula along with 6×4 mL dichloromethane washes of the treated product resin sample and the combined filtrates partitioned between 30 mL sat. aqueous sodium bicarbonate and 30 mL dichloromethane. The organic layer was isolated and the aqueous layer extracted with a further 5 6×30 mL diethyl ether. The organic extracts were then combined, dried over sodium sulfate, and concentrated in vacuo. Chromatographic purification of the resulting orange oil (SiO_2 , 3% methanol–dichloromethane on a column packed in eluent + 5% triethylamine) provided the expected cleaved amine substrate as a yellow-tinged clear oil (5.2 mg, 28%).

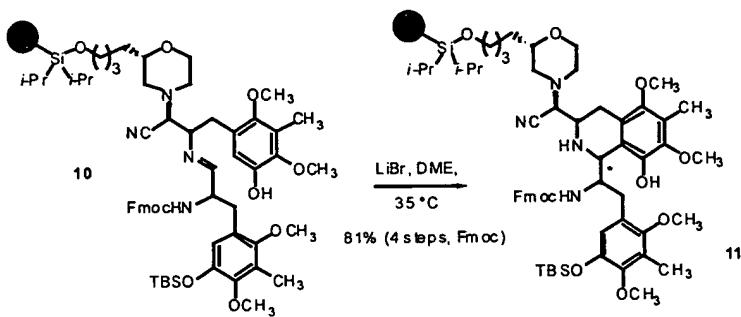
10 epi-9: ^1H NMR (400 MHz, CDCl_3), δ 6.61 (s, 1H, ArH), 3.91 (dm, 1H, J = 11.6 Hz, ROCH₂), 3.79 (s, 3H, ArOCH₃), 3.72–3.67 (m, 3H, ROCH₂, OCHCH₂, CHCN), 3.66 (s, 3H, ArOCH₃), 3.52–3.44 (m, 2H, HOCH₂), 3.32 (dt, 1H, J = 3.2, 8.4 Hz, CHNH₂), 3.16 (dd, 1H, J = 3.2, 13.2 Hz, NCH₂), 2.81 (d, 1H, J = 10.8 Hz, CH₂Ar), 2.65 (dt, 1H, J = 3.6, 11.2 Hz, NCH₂), 2.54 (d, 1H, J = 10.4 Hz, NCH₂), 2.37 (dd, 1H, J = 8.4, 13.6 Hz, NCH₂), 2.24 (s, 3H, ArCH₃), 2.22–15 2.15 (m, 1H, CH₂Ar), 1.64–1.40 (m, 6H, HOCH₂CH₂CH₂CH₂),. ^{13}C NMR (100 MHz, CDCl_3), δ 150.7, 145.5, 145.0, 127.2, 125.0, 116.4, 114.6, 75.7, 66.7, 65.9, 63.0, 60.9, 53.3, 52.8, 51.4, 35.2, 33.4, 32.8, 21.8, 17.4, 10.2. FTIR (neat film), cm^{-1} 3358 (m, NH₂/OH), 2936 (s), 2864 (m), 1483 (m), 1452 (m), 1417 (m), 1232 (w), 1112 (s), 1050 (m), 1009 (m). R_f 0.34, 10% methanol–dichloromethane, triethylamine-dipped plate. HRMS (TOF-ES⁺) Calcd for $\text{C}_{21}\text{H}_{34}\text{N}_3\text{O}_5$ ($\text{M}+\text{H}$)⁺:

20 408.2498, Found: 408.2480.



A solution of aldehyde Prepared as previously reported: (Myers, A. G.; Kung, D. W.; Zhong, B.; Movassaghi, M.; and Kwon, S. *J. Am. Chem. Soc.* 1999, 121, 8401–8402) (726.6 mg, 25 1.26 mmol, 2.84 equiv) in 15.0 mL N,N-dimethylformamide was added to amine resin **9** (1.96 g, 443.5 μ mol, 1.0 equiv, 0.2261 mmol/g) via cannula using 2×5 mL N,N-dimethylformamide

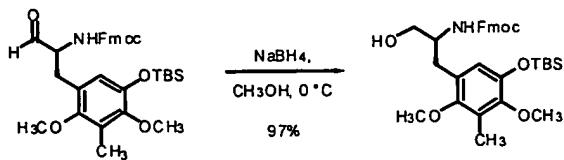
washes to quantitate the transfer. The resulting clear suspension of orangish resin was stirred at 23 °C in the dark for 5 hr before the reaction supernatant was removed via crimped cannula and the product resin washed sequentially with 2×20 mL N,N-dimethylformamide and 2×20 mL 1,2-dimethoxyethane. (Note: the reaction supernatant and N,N-dimethylformamide washes were collected to allow recovery of excess aldehyde (see below).) The resulting yellow resin (**10**) was employed directly in the synthesis of tetrahydroisoquinoline resin **11**.



A solution of anhydrous lithium bromide (3.41 g, 39.30 mmol, 88 equiv) in 30.0 mL 1,2-dimethoxyethane (prepared with gentle heating) was added to imine resin **10** (2.21 mg, 443.5 μ mol, 1.0 equiv, 0.2008 mmol/g) via a warm syringe and needle. The resulting clear suspension of yellow resin was placed on a 35 °C bath and stirred for 22 hr. The supernatant was then removed via crimped cannula and the product resin washed sequentially with 4 \times 20 mL N,N-dimethylformamide, 3 \times 20 mL tetrahydrofuran, and 2 \times 20 mL dichloromethane and dried in vacuo to provide free-flowing orangish-white resin which gave a negative Kaiser test and positive chloranil test. Photometric determination of the product resin 9-fluorenylmethoxycarbonyl (Fmoc) loading level (A_{290} , 20% piperidine-N,N-dimethylformamide, duplicate measurements) established a resin loading level of 0.1698 mmol/g, consistent with an 81% yield of diastereomeric tetrahydroisoquinolines (average 95% yield/step, 4 steps). Liberation of substrate for characterization and establishment of diastereomeric ratio for the transformation was effected by incubating a suspension of product resin **11** (86.8 mg, 13.2 μ mol (based on Fmoc loading level), 1.0 equiv) in 2.0 mL dichloromethane with methanol (53.5 μ L, 1.32 mmol, 100.0 equiv) and concentrated hydrochloric acid (5.4 μ L, 64.8 μ mol, 4.9 equiv) at 23 °C. After 1 hr, the reaction supernatant was collected via crimped cannula, along with 2 \times 2 mL dichloromethane washes of the treated resin sample. This filtrates were partitioned between 30

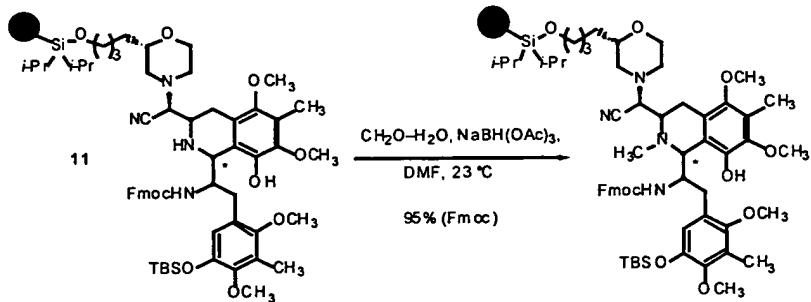
mL dichloromethane and 30 mL sat. aq. sodium bicarbonate, the organic layer isolated, and the aqueous layer extracted with a further 2×30 mL diethyl ether. The combined organic extracts were then dried over sodium sulfate, concentrated in vacuo, and chromatographically purified (SiO₂, 70% ethyl acetate–hexanes → 80% ethyl acetate–hexanes → 100% ethyl acetate–hexanes), affording *cis*-**11** (3.4 mg, 27%) and a mixture of *trans*-**11** and *epi*-**11** (epimeric at the aminonitrile methine and both centers in the tetrahydroisoquinoline ring) (0.8 mg, 6.3%, 1.8:1 *trans*- to *epi*-) as white foams (~6.7:1 diastereomeric ratio of *cis*- and *trans*- products).

cis-**11**: ¹H NMR (500 MHz, CDCl₃), * denotes non-fused aromatic ring protons, δ 7.74 (d, 2H, J = 8.0 Hz, ArH), 7.49 (d, 1H, J = 7.0 Hz, ArH), 7.43 (d, 1H, J = 7.5 Hz, ArH), 7.40–7.36 (m, 2H, ArH), 7.29–7.25 (m, 2H, ArH), 6.36 (s, 1H, ArH), 6.18 (s, 1H, ArOH), 5.68 (d, 1H, J = 6.5 Hz, NHFmoc), 4.82 (s, 1H, NHCH), 4.57 (br s, 1H, NCHAR), 4.43 (dd, 1H, J = 7.0, 10.5 Hz, CO₂CH₂), 4.16 (t, 1H, J = 7.5 Hz, CO₂CH₂CH), 4.09 (app. t, 1H, J = 10.0 Hz, CO₂CH₂), 3.93 (d, 1H, J = 10.5 Hz, ROCH₂CH₂), 3.79 (s, 3H, ArOCH₃*), 3.69 (s, 3H, ArOCH₃*), 3.65 (s, 6H, 2×ArOCH₃), 3.61–3.58 (m, 1H, ROCH₂CH₂), 3.57 (app. s, 2H, HOCH₂), 3.47 (d, 1H, J = 10.5 Hz, CHCN), 3.37 (br s, 1H, OCHCH₂), 3.20 (dd, 1H, J = 1.5, 14.5 Hz, CH₂Ar), 3.11 (t, 1H, J = 10.5 Hz, CH(CN)CH), 2.87 (t, 1H, J = 13.5 Hz, CHNHFmoc), 2.65 (t, 2H, J = 11.0 Hz, 2□NCH₂), 2.45 (t, 2H, J = 11.0 Hz, 2×NCH₂), 2.33 (dd, 1H, J = 11.0, 14.0 Hz, CH₂Ar), 2.25 (s, 3H, ArCH₃*), 2.19 (s, 3H, ArCH₃), 2.12 (t, 2H, J = 13.0 Hz, CH₂Ar*), 1.64–1.40 (m, 6H, HOCH₂CH₂CH₂CH₂), 0.96 (s, 9H, SiC(CH₃)₃), 0.10 (2×s, 6H, Si(CH₃)₂). ¹³C NMR (100 MHz, CDCl₃), δ 156.7, 149.3, 148.8, 145.9, 145.3, 144.5, 144.1, 144.0, 142.8, 141.5, 140.1, 127.9, 127.3, 125.4, 124.8, 122.6, 120.3, 120.2, 119.8, 115.0, 75.8, 67.1, 66.5, 64.2, 63.0, 60.9, 60.1, 58.3, 56.8, 55.2, 52.2, 50.5, 47.4, 47.2, 33.3, 32.9, 29.9, 29.2, 28.6, 25.9, 21.8, 18.4, 10.1, 9.9, –4.4. FTIR (neat film), cm^{–1} 3344 (m, NH), 2934 (s), 2857 (m), 1714 (s, NCO₂), 1463 (s), 1452 (s), 1238 (s), 1113 (s), 1060 (s), 869 (m), 783 (m). R_f 0.48, 80% ethyl acetate–hexanes. HRMS (TOF-ES⁺) Calcd for C₅₄H₇₃N₄O₁₀Si (M+H)⁺: 965.5096, Found: 965.5127.



The N,N-dimethylformamide solution of substrate aldehyde isolated by filtration of resin **10** (see above) was partitioned between 250 mL diethyl ether and 250 mL water and the ether layer isolated. The aqueous layer was extracted further with 2×250 mL (1:1) hexanes–diethyl ether and all organic extracts combined, dried over sodium sulfate, and concentrated in vacuo (some N,N-dimethylformamide remained). The resulting oil (471.2 mg aldehyde (theoretical), 818.5 μmol, 1.0 equiv) was dissolved in 27.0 mL methanol and stirred at 0 °C for 10 min before adding sodium borohydride in one portion (15.5 mg, 409.2 μmol, 0.5 equiv). After 25 min at 0 °C, the reaction solution was diluted with 15 mL diethyl ether and excess sodium borohydride carefully quenched by the SLOW addition of 15 mL saturated aqueous ammonium chloride. The resulting solution was partitioned between 150 mL diethyl ether and 150 mL saturated aqueous ammonium chloride and the organic layer separated and washed with 1×100 mL sat. aq. ammonium chloride and 1×100 mL brine. All combined aqueous washes were then extracted 1×250 mL diethyl ether and all organic extracts were then combined, dried over sodium sulfate, and concentrated in vacuo. Chromatographic purification of the resulting product (SiO_2 , 70% ethyl acetate–hexanes) provided the previously reported alcohol product (Myers, A. G.; Kung, D. W.; Zhong, B.; Movassaghi, M.; and Kwon, S. *J. Am. Chem. Soc.* **1999**, *121*, 8401–8402) as a clear oil (458.7 mg, 97%).

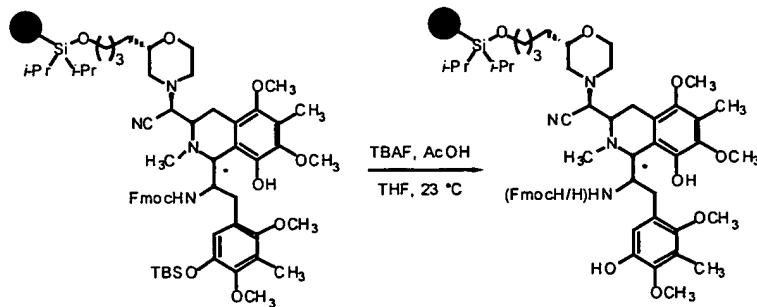
20



Formalin (660.5 μ L, 8.87 mmol, 20.0 equiv) was added to a suspension of tetrahydroisoquinoline resin **11** (2.21 mg, 443.5 μ mol, 1.0 equiv, 0.1762 mmol/g) and sodium triacetoxyborohydride (1.41 g, 6.65 mmol, 15.0 equiv) in 40.0 mL N,N-dimethylformamide. The resulting clear suspension of orangish resin was stirred for 3.5 hr before removing the 5 reaction supernatant via crimped cannula and washing the resin sequentially with 5 \times 30 mL N,N-dimethylformamide, 5 \times 30 mL tetrahydrofuran, and 2 \times 30 mL dichloromethane. Drying the product resin in vacuo provided a free-flowing, light orange resin than gave a negative chloranil test. Photometric determination of the product resin 9-fluorenylmethoxycarbonyl (Fmoc) loading level (A_{290} , 20% piperidine-N,N-dimethylformamide, duplicate measurements) 10 established a resin loading level of 0.1571 mmol/g indicating a 95% yield for this transformation given chromatographic and colorimetric evidence for the completion of this reaction. Substrate cleavage for characterization was effected by incubating product resin (95.5 mg, 16.8 μ mol, 1.0 equiv) in 2.7 mL dichloromethane with methanol (68.9 μ L, 1.70 mmol, 100.0 equiv) and concentrated hydrochloric acid (7.0 μ L, 83.3 μ mol, 4.9 equiv) for 18 hr at 23 °C. The reaction 15 supernatant was then removed by crimped cannula and collected along with 5 \times 2.5 mL dichloromethane washes of the treated resin. These filtrates were partitioned between 30 mL dichloromethane and 30 mL sat. aq. sodium bicarbonate and the organic layer was isolated. After further extraction of the aqueous layer (2 \times 30 mL diethyl ether), all organic extracts were combined, dried over sodium sulfate, and concentrated in vacuo. Chromatographic purification 20 of the resulting oil (prep TLC, SiO₂, 5% methanol–dichloromethane) provided the *cis*-tetrahydroisoquinoline product (10.2 mg, 62%) and *trans*-tetrahydroisoquinoline product (1.5 mg, 9%) as white solids (6.8:1 diastereomeric ratio).

cis-amine: ¹H NMR (500 MHz, CDCl₃), * denotes non-fused aromatic ring protons, δ 25 7.72 (dd, 2H, J = 4.0, 7.5 Hz, ArH), 7.41 (t, 2H, J = 7.5 Hz, ArH), 7.36 (t, 2H, J = 7.5 Hz, ArH), 7.26 (m, 2H, ArH), 6.52 (s, 1H, ArH), 6.17 (br s, 1H, ArOH), 5.50 (d, 1H, J = 8.5 Hz, NHFmoc), 4.17–4.14 (m, 1H, CO₂CH₂), 4.00 (t, 1H, J = 7.5 Hz, CO₂CH₂CH₂), 3.98–3.94 (m, 1H, CO₂CH₂), 3.93 (br s, 1H, NCHAR), 3.75 (d, 1H, J = 8.0 Hz, ROCH₂CH₂), 3.68 (2 \times s, 6H, 30 2 \times ArOCH₃*), 3.66 (s, 3H, ArOCH₃), 3.65 (s, 3H, ArOCH₃), 3.60 (s, 2H, HOCH₂), 3.54 (m, 1H, ROCH₂CH₂), 3.48 (dt, 1H, J = 2.0, 6.5 Hz, OCHCH₂), 3.43 (s, 1H, CHCN), 3.31 (d, 1H, J =

11.0 Hz, CH_2Ar), 2.96–2.93 (m, 1H, CHNCH_3), 2.93–2.88 (m, 1H, CHNHFmoc), 2.75–2.67 (m, 3H, 2 \square NCH_2 , CH_2Ar), 2.59 (s, 3H, NCH_3), 2.52 (app. dt, 2H, J = 2.5, 11.0 Hz, NCH_2 , CH_2Ar^*), 2.26–2.19 (m, 2H, NCH_2 , CH_2Ar^*), 2.19 (s, 3H, ArCH_3^*), 2.17 (s, 3H, ArCH_3), 1.61–1.49 (m, 4H, $\text{HOCH}_2\text{CH}_2\text{CH}_2\text{CH}_2$), 1.45–1.35 (m, 2H, $\text{HOCH}_2\text{CH}_2\text{CH}_2$), 0.95 (s, 9H, 5 $\text{Si}(\text{CH}_3)_3$), 0.11 (app. d, 6H, J = 5.5 Hz, $\text{Si}(\text{CH}_3)_2$). ^{13}C NMR (100 MHz, CDCl_3), δ 156.4, 151.6, 148.7, 148.5, 145.1, 144.3 (2), 144.1, 142.6, 141.3 (2), 127.8, 127.2 (2), 126.9, 125.4, 125.3, 125.2, 123.7, 122.9, 120.6, 120.1, 120.0, 116.3, 75.8, 66.9, 66.6, 64.7, 63.9, 63.7, 63.0, 61.4, 61.2, 60.8, 60.0, 58.4, 57.9, 50.5, 47.4, 33.3, 32.8, 31.2, 29.9, 25.9, 24.7, 21.8, 18.4, 10.1, 9.8, –4.4. FTIR (neat film), cm^{-1} 3390 (m, NH), 2932 (s), 2857 (m), 1708 (s, NCO_2), 1480 (m), 10 1450 (m), 1416 (m), 1237 (m), 1115 (m), 1061 (s), 1009 (m), 870 (m), 741 (m). R_f 0.42, 5% methanol–dichloromethane. HRMS (TOF-ES $^+$) Calcd for $\text{C}_{55}\text{H}_{75}\text{N}_4\text{O}_{10}\text{Si}$ ($\text{M}+\text{H}$) $^+$: 979.5252, Found: 979.5210.

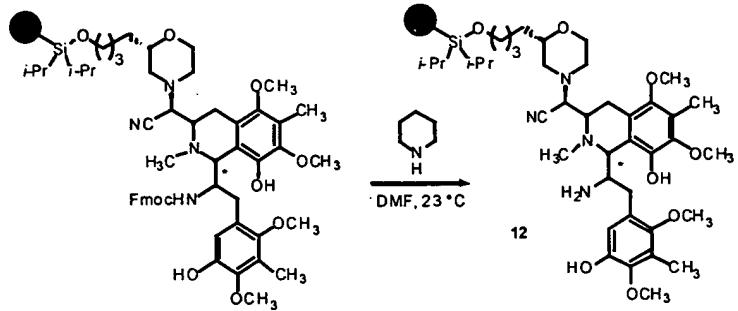


15

Tetra-*n*-butylammonium fluoride (1.0 M in tetrahydrofuran, 2.03 mL, 2.03 mmol, 5.0 equiv) was added to a suspension of N-methylated amine resin (2.42 g, 405.4 μmol , 1.0 equiv, 0.1678 mmol/g) and glacial acetic acid (231.7 μL , 4.05 mmol, 10.0 equiv) in 20.0 mL tetrahydrofuran at 23 °C. The resulting suspension was stirred for 2.5 hr before the reaction 20 supernatant was removed via crimped cannula. The product resin was washed sequentially with 2×20 mL N,N-dimethylformamide, 3×20 mL tetrahydrofuran, and 2×20 mL dichloromethane and dried in vacuo to provide a free-flowing yellow-orange resin. Cleaved substrate was recovered from the isolated reaction supernatant and N,N-dimethylformamide washes to determine the extent of siloxane cleavage through this procedure. The washes and supernatant 25 were concentrated in vacuo to ~3 mL then partitioned between 30 mL diethyl ether and 30 mL (3:1) water–sodium bicarbonate (sat., aq.). The organic layer was separated and the aqueous

layer extracted with 2×30 mL diethyl ether. All organic extracts were combined, dried over sodium sulfate, concentrated in vacuo, and chromatographically purified (prep TLC, 7% methanol–dichloromethane) to provide the expected siloxane cleavage products, the *cis*-tetrahydroisoquinoline (4.5 mg, 1.3%), and presumed *trans*-tetrahydroisoquinoline (0.5 mg, ~0.14%), as white solids. Photometric determination of the untreated product resin 9-fluorenylmethoxycarbonyl (Fmoc) loading level (A_{290} , 20% piperidine–N,N-dimethylformamide, duplicate measurements) established a resin loading level of 0.07732 mmol/g, suggesting that ~58% of the resin-bound product had lost its Fmoc protecting group in this transformation (assuming a 98% yield for the phenolic siloxane deprotection (based on substrate resin-cleavage yields)). This deprotection of resin-bound product was confirmed by the isolation of dibenzofulvene (13.2 mg, 33%) from the reaction supernatant along with the diastereomeric tetrahydroisoquinolines described above.

cis-tetrahydroisoquinoline: ^1H NMR (500 MHz, CDCl_3), * denotes non-fused aromatic ring protons, δ 7.72 (d, 2H, J = 7.2 Hz, ArH), 7.43 (d, 2H, J = 8.0 Hz, ArH), 7.36 (q, 2H, J = 7.2 Hz, ArH), 7.26 (app. q, 2H, ArH), 6.64 (s, 1H, ArH), 6.21 (br s, 1H, ArOH), 5.63 (d, 1H, J = 8.4 Hz, NHFmoc), 5.34 (br s, 1H, ArOH*), 4.24 (dd, 1H, J = 6.8, 10.4 Hz, CO_2CH_2), 4.16 (d, 1H, J = 6.0 Hz, CO_2CH_2), 4.03 (t, 1H, J = 7.2 Hz, $\text{CO}_2\text{CH}_2\text{CH}$), 3.98–3.89 (m, 2H, NCHAR, ROCH_2CH_2), 3.74 (d, 1H, J = 4.8 Hz, ROCH_2CH_2), 3.70 (s, 3H, ArOCH₃*), 3.69 (s, 3H, ArOCH₃*), 3.68 (s, 3H, ArOCH₃), 3.66 (s, 3H, ArOCH₃), 3.62 (m, 1H, OCHCH₂), 3.54 (br s, 2H, HOCH₂), 3.48 (m, 1H, CHCN), 3.31 (d, 1H, J = 11.6 Hz, CH₂Ar), 3.03 (dd, 1H, J = 10.8, 13.6 Hz, CHNCH₂), 2.93 (d, 1H, J = 12.0 Hz, CHNHFmoc), 2.83 (dd, 1H, J = 11.2 Hz, CH₂Ar), 2.75–2.66 (m, 2H, 2×NCH₂), 2.59 (s, 3H, NCH₃), 2.53 (dt, 2H, J = 2.8, 12.4 Hz, NCH₂, CH₂Ar*), 2.26–2.18 (m, 2H, NCH₂, CH₂Ar*), 2.23 (s, 3H, ArCH₂*), 2.18 (s, 3H, ArCH₃), 1.62–1.38 (m, 6H, HOCH₂CH₂CH₂CH₂). ^{13}C NMR (100 MHz, CDCl_3), δ 156.5, 150.7, 148.5, 145.3, 144.5, 144.3, 144.1, 142.6, 141.4, 128.1, 127.8, 127.2 (2), 125.5, 125.3, 124.2, 123.6, 123.0, 120.5, 120.1, 116.4, 114.5, 75.8, 66.9, 66.6, 64.5, 63.6, 63.0, 61.4, 61.1, 61.0, 60.9 (2), 58.4, 58.2, 50.6, 47.4, 47.3, 33.3, 32.8, 31.3, 29.9, 24.7, 21.7, 10.2, 9.8. FTIR (neat film), cm^{-1} 3392 (m, NH), 2939 (s), 2861 (m), 1702 (s, NCO₂), 1451 (m), 1416 (m), 1233 (m), 1112 (m), 1052 (s), 1009 (m), 910 (m), 733 (s). R_f 0.25, 90% ethyl acetate–hexanes. HRMS (TOF-ES⁺) Calcd for $\text{C}_{49}\text{H}_{61}\text{N}_4\text{O}_{10}$ ($\text{M}+\text{H}$)⁺: 865.4387, Found: 865.4390.

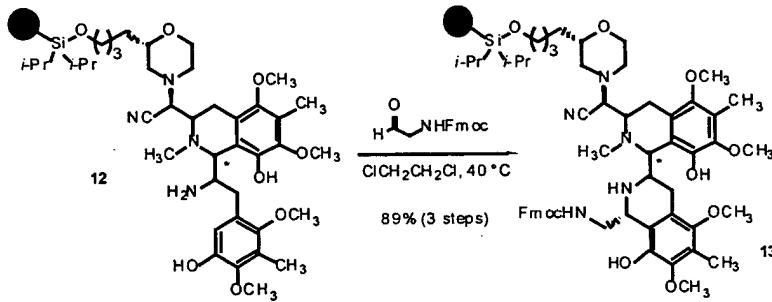


5 Substrate phenol resin (2.19 g, 355.5 μmol , 1.0 equiv, 0.1681 mmol/g) was suspended in
 20.0 mL 20% (v/v) piperidine-N,N-dimethylformamide. The resulting clear suspension of
 orange resin was stirred for 1.5 hr at 23 °C before removing the reaction supernatant via crimped
 cannula. The product resin was sequentially washed with 3×20 mL N,N-dimethylformamide,
 3×20 mL tetrahydrofuran, and 2×20 mL dichloromethane and dried in vacuo to provide a free-
 flowing orange resin. Soluble substrate was isolated for characterization by incubating a sample
 10 of product resin 12 (74.3 mg, 12.53 μmol , 1.0 equiv) in 2.0 mL dichloromethane with methanol
 (50.8 μL , 1.25 mmol, 100.0 equiv) and concentrated hydrochloric acid (5.1 μL , 61.42 μmol , 4.9
 equiv) for 18.5 hr at 23 °C. The reaction supernatant from this mixture was then collected via
 15 crimped cannula along with 6×2 mL dichloromethane washes of the treated resin. The combined
 washes and supernatant were partitioned between 30 mL dichloromethane and 30 mL sat. aq.
 sodium bicarbonate and the organic layer isolated. After further extraction of the aqueous layer
 (2×20 mL diethyl ether), all organic extracts were combined, dried over sodium sulfate, and
 concentrated in vacuo. Chromatographic purification of the resulting oil (SiO_2 , 2% methanol-
 20 dichloromethane, column packed with eluent + 1% triethylamine) afforded the expected *cis*-
cis-tetrahydroisoquinoline product as a white solid (1.6 mg, 20%).

***cis*-12:** ^1H NMR (400 MHz, CDCl_3), * denotes non-fused aromatic ring protons, δ 6.78
 (s, 1H, ArH), 6.66 (s, 1H, ArOH), 5.60 (s, 1H, ArOH*), 3.98 (br s, 1H, NCHAR), 3.94 (d, 1H, J
 = 10.8 Hz, ROCH₂CH₂), 3.82 (s, 3H, ArOCH₃*), 3.77 (s, 3H, ArOCH₃*), 3.73 (m, 1H,
 25 ROCH₂CH₂), 3.68 (s, 3H, ArOCH₃), 3.66 (s, 3H, ArOCH₃), 3.64 (t, 2H, J = 6.0 Hz, HOCH₂),
 3.54 (m, 2H, OCHCH₂, CHCN), 3.32 (dd, 1H, J = 4.4, 15.2 Hz, CH₂Ar), 3.17 (m, 1H, NCH₂),

2.92 (d, 1H, J = 10.8 Hz, NCH_2), 2.79–2.71 (m, 3H, CHNCH_3 , CHNH_2 , NCH_2), 2.62 (s, 3H, NCH_3), 2.52 (dt, 1H, J = 2.8, 10.8 Hz, NCH_2), 2.41 (dd, 1H, J = 13.2, 15.2 Hz, CH_2Ar), 2.24–2.20 (m, 2H, 2 \times CH_2Ar^*), 2.22 (s, 3H, ArCH_3^*), 2.21 (s, 3H, ArCH_3), 1.60–1.38 (m, 6H, $\text{HOCH}_2\text{CH}_2\text{CH}_2\text{CH}_2$). ^{13}C NMR (100 MHz, CDCl_3), δ 150.4, 148.7, 146.4, 145.9, 145.0, 143.7, 5 124.8, 124.1, 123.4, 122.3, 116.6, 114.8, 75.7, 66.5, 64.9, 62.9, 61.7, 61.4, 61.1, 60.9, 60.8, 59.9, 59.7, 59.1, 58.2, 50.7, 47.3, 33.2, 32.7, 23.9, 21.7, 10.2, 9.8. FTIR (neat film), cm^{-1} 3354 (w, OH/NH), 2936 (m), 2859 (w), 1455 (m), 1416 (m), 1112 (s), 1053 (m), 1005 (m). R_f 0.49, 10% methanol–dichloromethane, triethylamine-dipped plate. HRMS (TOF-ES $^+$) Calcd for $\text{C}_{34}\text{H}_{51}\text{N}_4\text{O}_8$ ($\text{M}+\text{H}$) $^+$: 643.3707, Found: 643.3713.

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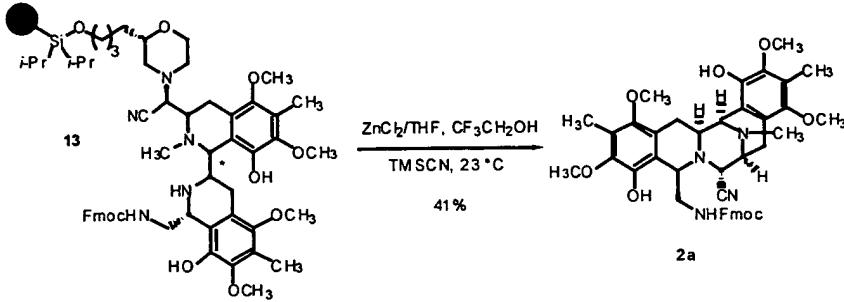
A solution of *N*-(9-fluorenylmethoxycarbonyl)-glycinal (Prepared as reported in Myers, A. G.; Kung, D. W. *J. Am. Chem. Soc.* **1999**, *121*, 10828–10829) (25.8 mg, 91.50 μmol , 3.0 equiv) in 3.1 mL dichloroethane was freeze-pump-thaw deoxygenated (3 cycles) and added via cannula to amine resin **12** (180.8 mg, 30.50 μmol , 1.0 equiv). The resulting resin suspension was stirred at 40 °C for 20 hr before removing the reaction supernatant via crimped cannula. The product resin was then washed with 6 \times 3 mL tetrahydrofuran and 3 \times 3 mL dichloromethane and dried in vacuo, yielding a free-flowing orange resin. Photometric determination of the product resin 9-fluorenylmethoxycarbonyl (Fmoc) loading level (A_{290} , 20% piperidine–N,N-dimethylformamide, duplicate measurements) established an Fmoc-loading level of 0.2076 mmol/g, consistent with a yield of >100% for this transformation, suggesting that product resin **13** has the ability to bind extra equivalents of aldehyde. An isolated yield for this transformation was obtained by incubating product resin **13** (88.58 mg, 14.31 μmol , 1.0 equiv) in 2.3 mL dichloromethane with methanol (57.9 μL , 1.43 mmol, 100.0 equiv) and concentrated hydrochloric acid (4.8 μL , 57.2 μmol , 4.9 equiv) for 19 hr at 23 °C. The reaction supernatant 15

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was then collected via crimped cannula along with 6×2 mL dichloromethane washes of the treated resin and the combined filtrates partitioned between 30 mL dichloromethane and 30 mL sat. aq. sodium bicarbonate. The organic layer was isolated and the aqueous layer further extracted with 2×30 mL diethyl ether. All organic extracts were then combined, dried over sodium sulfate, and concentrated in vacuo to provide an orange oil. Chromatographic purification of this oil (prep TLC, SiO₂, 10% methanol–dichloromethane) afforded the expected *cis*-/*cis*-bis-tetrahydroisoquinoline cleavage product (5.7 mg, 44%, 3 steps) as a yellow-orange solid along with several diastereomeric products (various centers, 5.8 mg, 45%, 3 steps) that were not cleanly separated by prep TLC. The combined 89% isolated yield over three steps corresponds to an average yield per step of ~96%.

cis-13: ¹H NMR (500 MHz, CDCl₃), * denotes unmethylated tetrahydroisoquinoline ring system, δ 7.76 (dd, 2H, J = 3.5, 8.0 Hz, ArH), 7.59–7.54 (m, 2H, ArH), 7.41–7.38 (m, 2H, ArH), 7.30 (q, 2H, J = 7.0 Hz, ArH), 5.72 (br s, 1H, ArOH), 5.37 (br s, 1H, NHFmoc), 4.37 (d, 2H, J = 7.5 Hz, CO₂CH₂, NCHAR), 4.34 (d, 1H, CO₂CH₂), 4.21 (t, 1H, J = 6.5 Hz, CO₂CH₂CH), 3.91 (d, 2H, J = 11.5 Hz, ROCH₂CH₂, NCHAR*), 3.76 (s, 3H, ArOCH₃*), 3.74 (s, 3H, ArOCH₃*), 3.71 (m, 1H, ROCH₂CH₂), 3.68 (m, 1H, OCHCH₂), 3.66 (2×s, 6H, 2×ArOCH₃), 3.64 (s, 1H, CHCN), 3.49 (m, 4H, HOCH₂, CH₂NHFmoc), 3.27 (d, 1H, J = 11.5 Hz, CH₂Ar), 2.84 (d, 2H, J = 11.0 Hz, NCH₂, CH₂Ar*), 2.71–2.68 (br s, 3H, NCH₃), 2.67–2.56 (m, 2H, CHNCH₃, CHNH), 2.48 (dt, 2H, J = 3.5, 11.0 Hz, 2×NCH₂), 2.42 (m, 1H, CH₂Ar), 2.24–2.21 (m, 2H, NCH₂, CH₂Ar*), 2.22 (s, 3H, ArCH₃*), 2.21 (s, 3H, ArCH₃), 1.61–1.54 (m, 2H, HOCH₂CH₂), 1.54–1.46 (m 2H, HOCH₂CH₂CH₂CH₂), 1.46–1.34 (m, 2H, HOCH₂CH₂CH₂). ¹³C NMR (100 MHz, CDCl₃), δ 157.5, 149.4, 148.7, 145.7, 144.3, 143.9, 143.7, 142.0, 141.5, 131.1, 130.8, 128.6, 127.9, 127.3, 126.2, 125.4, 123.7, 122.6, 120.2, 116.3, 75.8, 67.1, 66.5, 64.9, 63.0, 61.4, 61.1, 61.0, 60.7, 59.1, 58.3, 52.8, 49.9, 47.4, 47.0, 46.6, 33.3, 32.8, 29.9, 27.2, 24.2, 21.8, 9.9, 9.8. FTIR (neat film), cm⁻¹ 3390 (m, NH/OH), 2936 (s), 2863 (m), 1713 (m, NCO₂), 1451 (s), 1413 (m), 1258 (m), 1112 (s), 1057 (s), 1006 (m), 734 (s). R_f 0.34, 10% methanol–dichloromethane. HRMS (TOF-ES⁺) Calcd for C₅₁H₆₄N₅O₁₀ (M+H)⁺: 906.4653, Found: 906.4688.



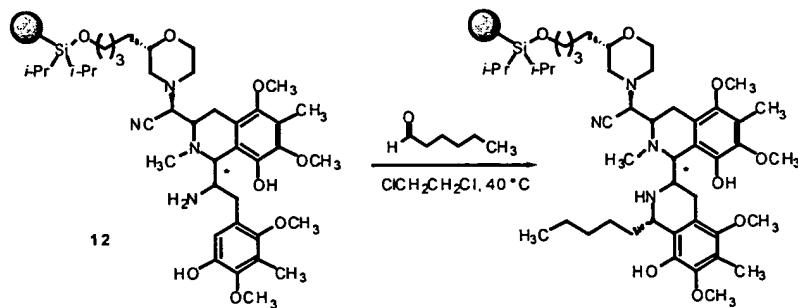
2,2,2-trifluoroethanol (35.2 μ L, 483.10 μ mol, 68.6 equiv), zinc chloride (0.5 M in tetrahydrofuran, 70.4 μ L, 35.21 μ mol, 5.0 equiv), and trimethylsilylcyanide (3.8 μ L, 28.17 μ mol, 5.0 equiv) were sequentially added to a suspension of resin **13** (43.6 mg, 7.04 μ mol, 1.0 equiv) in 105.6 μ L tetrahydrofuran at 23 $^{\circ}$ C. The resulting yellow suspension was stirred for 16 hr at 23 $^{\circ}$ C before diluting the reaction mixture with 1.5 mL tetrahydrofuran. The reaction supernatant was then collected via crimped cannula along with 5 \times 2 mL tetrahydrofuran washes of the treated resin. The collected filtrates were partitioned between 5 mL ethyl acetate and 15 mL of an aqueous solution of 0.2 N ethylenediaminetetraacetic acid disodium salt dihydrate and 0.4 N sodium hydroxide (pH 10). The organic layer was isolated and washed with 1 \times 10 mL brine and the aqueous layers then combined and extracted with a further 2 \times 15 mL ethyl acetate. The combined organic extracts were then dried over sodium sulfate and concentrated in vacuo. Chromatographic purification of the resulting red oil (prep TLC, SiO₂, 5% methanol–dichloromethane) afforded pentacyclic product **2a** as a white solid (2.0 mg, 41%) along with small amounts of the product aminonitrile hydrolysis product (0.4 mg, 8%).

2a: ¹H NMR (400 MHz, CDCl₃), δ 7.76 (t, 2H, J = 6.8 Hz, ArH), 7.47–7.39 (m, 2H, ArH), 7.30 (t, 2H, J = 7.2 Hz, ArH), 5.57 (br s, 1H, ArOH), 5.51 (br s, 1H, ArOH), 4.53 (t, 1H, J = 6.0 Hz, NHFmoc), 4.32 (dd, 1H, J = 6.8, 10.8 Hz, CO₂CH₂), 4.22 (dd, 1H, J = 6.0, 10.8 Hz, CO₂CH₂), 4.14 (br s, 1H, ArCHNCH₃), 4.11 (t, 1H, J = 4.4 Hz, ArCHCH₂NHFmoc), 4.06 (t, 1H, J = 6.4 Hz, CO₂CH₂CH), 3.74 (s, 3H, ArOCH₃), 3.60 (s, 6H, 2 \times ArOCH₂), 3.55 (s, 3H, ArOCH₃), 3.29 (d, 1H, J = 7.2, ArCH₂CHNCH₃), 3.24–3.19 (m, 3H, ArCH₂CHNC(CN), ArCH₂CHNC(CN), CH₂NHFmoc), 3.11–3.04 (m, 1H, CH₂NHFmoc), 2.96 (dd, 1H, J = 8.0, 18.8 Hz, ArCH₂CHNCH₃), 2.33 (d, 1H, J = 11.6 Hz, ArCH₂CHNCH₃), 2.31 (s, 3H, NCH₃), 2.19 (s, 3H, ArCH₃), 2.11 (s, 3H, ArCH₃), 1.88 (dd, 1H, J = 11.6, 16.4 Hz, ArCH₂CHNC(CN)). R_f 0.38,

5% methanol–dichloromethane. LRMS (TOF-ES⁺) Calcd for C₄₃H₄₇N₄O₈ (M+H)⁺: 747, Found: 747.

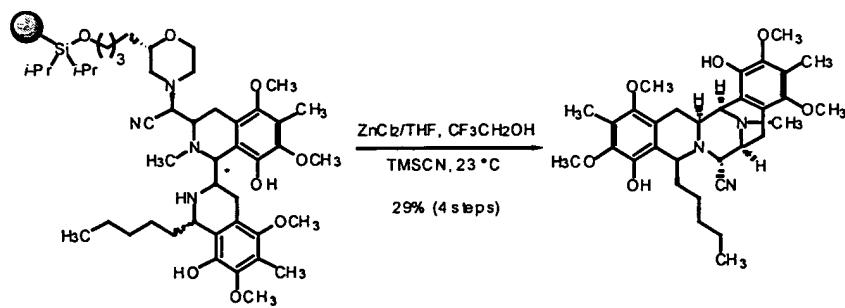
Exemplary solid-supported syntheses of compounds having alternate pentacyclic core

5 structures:



A solution of hexanal (7.8 μ L, 64.70 μ mol, 5.0 equiv) in 1.3 mL dichloroethane was
10 freeze-pump-thaw deoxygenated (3 cycles) and added via cannula to amine resin 12 (76.7 mg,
12.94 μ mol, 1.0 equiv). The resulting resin suspension was stirred in the dark at 40 °C for 20.5
hr before removing the reaction supernatant via crimped cannula. The product resin was then
washed with 6×3 mL tetrahydrofuran and 3×3 mL dichloromethane and dried in vacuo, yielding
a free-flowing orange resin. The product resin was directly employed in subsequent reactions.

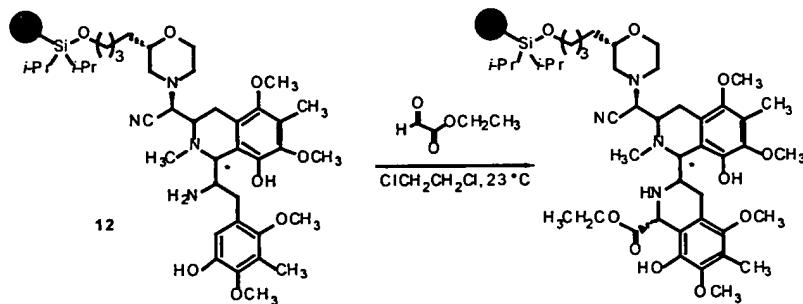
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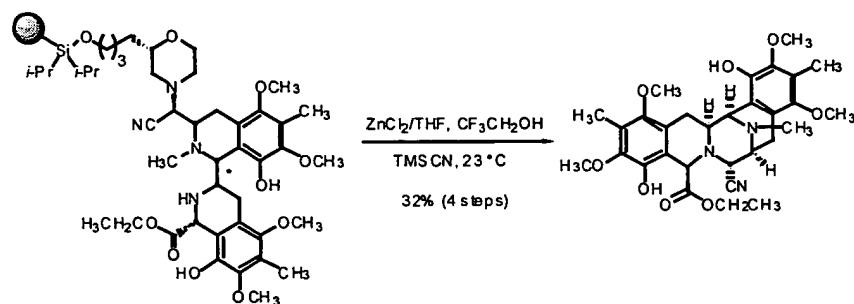
Zinc chloride (0.5 M in tetrahydrofuran, 2.3 mL, 1.16 mmol, 100.0 equiv) and trimethylsilylcyanide (151.8 μ L, 1.14 mmol, 99.0 equiv) were sequentially added to a suspension
20 of substrate resin (69.8 mg, 11.61 μ mol, 1.0 equiv) in 460.0 μ L 2,2,2-trifluoroethanol at 23 °C.
The resulting yellow suspension was stirred for 15.5 hr at 23 °C before removing and collecting

the reaction supernatant via crimped cannula along with 5×3 mL tetrahydrofuran washes of the treated resin. The collected filtrates were partitioned between 10 mL ethyl acetate and 20 mL of an aqueous solution of 0.2 N ethylenediaminetetraacetic acid disodium salt dihydrate and 0.4 N sodium hydroxide (pH 10). The organic layer was isolated and washed with 1×15 mL brine and 5 the aqueous layers then combined and extracted with a further 2×15 mL ethyl acetate. The combined organic extracts were then dried over sodium sulfate and concentrated in vacuo. Chromatographic purification of the resulting red oil (SiO_2 , 40% ethyl acetate–hexanes) afforded the pentacyclic product as a white solid (1.9 mg, 29%, 4 steps).

10 ^1H NMR (500 MHz, C_6D_6), ~2:1 mixture of rotamers, * denotes minor rotamer signals, δ 5.75 (s, 1H, ArOH), 5.18 (s, 1H, ArOH*), 5.17 (s, 1H, ArOH), 4.20 (m, 1H, ArCHNCH₃), 4.16 (d, 1H, J = 2.0 Hz, ArCHNCH₃*), 3.93 (m, 1H, ArCHNC(CN)*), 3.90 (m, 1H, ArCHNC(CN)), 3.83 (d, 1H, J = 3.2 Hz, CHCN), 3.73 (d, 1H, CHCN*), 3.53 (s, 3H, ArOCH₃*), 3.38 (s, 3H, ArOCH₃), 3.31 (s, 3H, ArOCH₃), 3.22 (s, 3H, ArOCH₃*), 3.14 (s, 3H, ArOCH₃*), 3.08 (s, 3H, ArOCH₃), 3.02 (s, 3H, ArOCH₃), 3.00 (s, 3H, ArOCH₃*), 2.98 (m, 1H, ArCH₂CHNCH₃), 2.80 (d, 1H, J = 13.4 Hz, ArCH₂CHNC(CN)), 2.66 (dd, 1H, J = 8.0, 18.8 Hz, ArCH₂CHNC(CN)), 15 2.58 (dt, 1H, J = 4.4, 12.8 Hz, ArCH₂CHNCH₃), 2.23 (dt, 1H, 4.4, 18.0 Hz, ArCH₂CHNCH₃), 2.15 (s, 3H, NCH₃), 2.13 (m, 1H, ArCH₂CHNC(CN)), 2.12 (s, 3H, ArCH₃*), 2.11 (s, 3H, ArCH₃), 2.05 (s, 3H, ArCH₃), 2.04 (m, 1H, ArCHCH₂), 1.95 (d, 1H, J = 18.8 Hz, ArCHCH₂), 20 1.62 (dt, 2H, J = 4.8, 13.2 Hz, ArCHCH₂CH₂), 0.90–0.78 (m, 4H, CH₂CH₂CH₂CH₃), 0.65 (t, 3H, J = 7.2 Hz, CH₂CH₃). FTIR (neat film), cm^{-1} 2934 (m), 2859 (w), 1462 (s), 1414 (s), 1280 (m), 1109 (m), 1063 (m), 1005 (m). R_f 0.32, 5% methanol–dichloromethane.



A solution of ethyl glyoxylate (50% w/w in toluene, 13.0 μ L, 65.79 μ mol, 5.0 equiv) in 1.3 mL dichloroethane was freeze-pump-thaw deoxygenated (3 cycles) and added via cannula to amine resin **12** (78.0 mg, 13.16 μ mol, 1.0 equiv). The resulting resin suspension was stirred in the dark at 23 °C for 22 hr before removing the reaction supernatant via crimped cannula. The product resin was then washed with 2 \times 3 N,N-dimethylformamide, 4 \times 3 mL tetrahydrofuran, and 2 \times 2 mL dichloromethane and dried in vacuo, yielding a free-flowing orange resin. The product resin was directly employed in subsequent reactions.

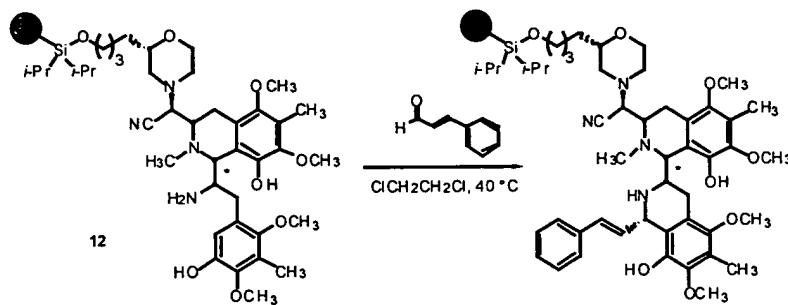


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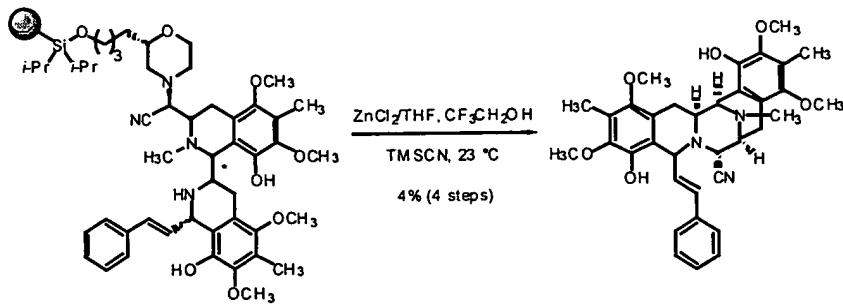
2,2,2-trifluoroethanol (61.6 μ L, 845.4 μ mol, 68.6 equiv), zinc chloride (0.5 M in tetrahydrofuran, 123.3 μ L, 61.64 μ mol, 5.0 equiv), and trimethylsilylcyanide (6.6 μ L, 49.31 μ mol, 4.0 equiv) were sequentially added to a suspension of substrate resin (74.1 mg, 12.33 μ mol, 1.0 equiv) in 185.0 μ L tetrahydrofuran at 23 °C. The resulting yellow suspension was stirred for 7 hr at 23 °C before diluting the reaction mixture with 1.5 mL tetrahydrofuran. The reaction supernatant was then collected via crimped cannula along with 5 \times 2 mL tetrahydrofuran washes of the treated resin. The collected filtrates were partitioned between 5 mL ethyl acetate and 15 mL of an aqueous solution of 0.2 N ethylenediaminetetraacetic acid disodium salt dihydrate and 0.4 N sodium hydroxide (pH 10). The organic layer was isolated and washed with 1 \times 10 mL brine and the aqueous layers then combined and extracted with a further 2 \times 15 mL ethyl acetate. The combined organic extracts were then dried over sodium sulfate and concentrated in vacuo. Chromatographic purification of the resulting orange oil (prep TLC, SiO₂, 5% methanol–dichloromethane) afforded the pentacyclic product as a yellowish-white solid (2.2 mg, 31.5%, 4 steps)

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¹H NMR (400 MHz, CDCl₃), δ 5.56 (s, 1H, ArOH), 5.51 (s, 1H, ArOH), 4.57 (s, 1H, ArCHCO₂), 4.35 (d, 1H, J = 2.4 Hz, CHCN), 4.17 (dd, 1H, J = 1.2, 2.8 Hz, ArCHNCH₃), 4.04–3.97 (m, 1H, CO₂CH₂), 3.95–3.85 (m, 1H, CO₂CH₂), 3.75 (s, 3H, ArOCH₃), 3.74 (s, 3H, ArOCH₃), 3.65 (s, 3H, ArOCH₃), 3.63 (s, 3H, ArOCH₃), 3.39 (d, 1H, J = 8.0 Hz, 5 ArCH₂CHC(CN)), 3.26 (dt, 1H, J = 2.4, 11.2 Hz, ArCH₂CHNC(CN)), 3.22 (dd, 1H, J = 2.8, 15.6 Hz, ArCH₂CHNC(CN)), 3.03 (dd, 1H, J = 8.0, 18 Hz, ArCH₂CHNCH₃), 2.41 (d, 1H, J = 18.4 Hz, ArCH₂CHNCH₃), 2.30 (s, 3H, NCH₃), 2.21 (s, 3H, ArCH₃), 2.20 (s, 3H, ArCH₃), 2.03 (dd, 1H, J = 11.6, 15.2 Hz, ArCH₂CHNC(CN)), 1.01 (t, 3H, J = 7.2 Hz, CO₂CH₂CH₃). FTIR (neat film), cm⁻¹ 3422 (m, OH), 2936 (m), 2829 (w), 1728 (m, CO₂), 1464 (s), 1414 (s), 1276 (m), 1052 (m), 1061 (s), 1027 (m), 1004 (m). R_f 0.39, 5% methanol–dichloromethane. HRMS (TOF-ES⁺) Calcd for C₃₀H₃₈N₃O₈ (M+H)⁺: 568.2658, Found: 568.2677.



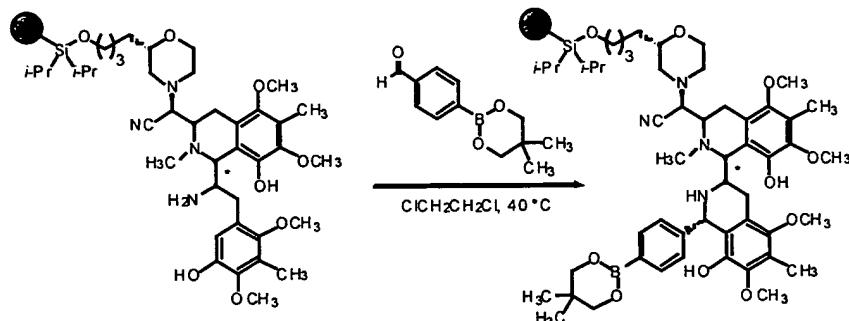
15 A solution of *trans*-cinnamaldehyde (8.8 μL, 69.42 μmol, 5.0 equiv) in 1.35 mL dichloroethane was freeze-pump-thaw deoxygenated (3 cycles) and added via cannula to amine resin 12 (82.3 mg, 13.88 μmol, 1.0 equiv). The resulting resin suspension was stirred in the dark at 40 °C for 21 hr before removing the reaction supernatant via crimped cannula. The product resin was then washed with 2×3 N,N-dimethylformamide, 4×3 mL tetrahydrofuran, and 2×2 mL 20 dichloromethane and dried in vacuo, yielding a free-flowing orange resin. The product resin was directly employed in subsequent reactions.



2,2,2-trifluoroethanol (65.0 μ L, 892.1 μ mol, 68.3 equiv), zinc chloride (0.5 M in tetrahydrofuran, 130.6 μ L, 65.29 μ mol, 5.0 equiv), and trimethylsilylcyanide (7.0 μ L, 52.23 μ mol, 4.0 equiv) were sequentially added to a suspension of substrate resin (78.9 mg, 13.06 μ mol, 1.0 equiv) in 196.0 μ L tetrahydrofuran at 23 °C. The resulting yellow suspension was stirred for 5.5 hr at 23 °C before diluting the reaction mixture with 1.5 mL tetrahydrofuran. The reaction supernatant was then collected via crimped cannula along with 5 \times 2 mL tetrahydrofuran washes of the treated resin. The collected filtrates were partitioned between 5 mL ethyl acetate and 15 mL of an aqueous solution of 0.2 N ethylenediaminetetraacetic acid disodium salt dihydrate and 0.4 N sodium hydroxide (pH 10). The organic layer was isolated and washed with 1 \times 10 mL brine and the aqueous layers then combined and extracted with a further 2 \times 15 mL ethyl acetate. The combined organic extracts were then dried over sodium sulfate and concentrated in vacuo. Chromatographic purification of the resulting yellow-brown oil (prep TLC, SiO₂, 5% methanol–dichloromethane) afforded the pentacyclic product as a white solid (0.3 mg, 4%, 4 steps)

¹H NMR (400 MHz, CDCl₃), δ 7.21–7.12 (m, 5H, ArH), 6.42 (d, 1H, J = 15.2 Hz, CHCHAr), 6.03 (dd, 1H, J = 5.6, 15.6 Hz, CHCHAr), 5.56 (s, 1H, ArOH), 5.49 (s, 1H, ArOH), 4.59 (d, 1H, J = 6.0 Hz, ArCHCHCHAr), 4.21 (d, 1H, J = 2.4 Hz, CHCN), 4.02 (d, 1H, J = 2.8 Hz, ArCHNCH₃), 3.78 (s, 3H, ArOCH₃), 3.73 (s, 3H, ArOCH₃), 3.61 (s, 3H, ArOCH₃), 3.60 (s, 3H, ArOCH₃), 3.39 (d, 1H, J = 6.4 Hz, ArCH₂CHC(CN)), 3.32 (dt, 1H, J = 12.0 Hz, ArCH₂CHNC(CN)), 3.26 (dd, 1H, J = 2.4, 15.6 Hz, ArCH₂CHNC(CN)), 3.04 (dd, 1H, J = 8.0, 18.4 Hz, ArCH₂CHNCH₃), 2.48 (d, 1H, J = 18.8 Hz, ArCH₂CHNCH₃), 2.35 (s, 3H, NCH₃), 2.28 (s, 3H, ArCH₃), 2.18 (s, 3H, ArCH₃), 2.00 (dd, 1H, J = 12.8, 14.0 Hz, ArCH₂CHNC(CN)). FTIR (neat film), cm⁻¹ 3357 (w, OH), 2918 (m), 2849 (w), 1463 (s), 1415 (s), 1109 (s), 1060 (s),

1002 (s), 744 (m). R_f 0.37, 5% methanol–dichloromethane. HRMS (ApCl⁺) Calcd for C₃₄H₃₉N₂O₆ (M-CN)⁺: 571, Found: 571.

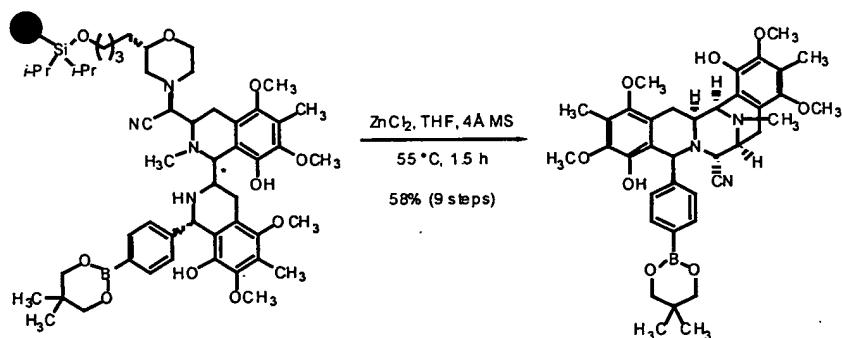


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A solution of benzaldehyde boronate ester (5.2 mg, 23.87 μ mol, 5.0 equiv) in 600 μ L 1,2-dichloroethane was freeze-pump-thaw deoxygenated (3 cycles) and then added via cannula to the amine resin (28.3 mg, 4.77 μ mol, 0.1687 mmol/g, 1.0 equiv). The resulting suspension 10 was stirred at 75 °C for 31 h and the supernatant was then removed via cannula. The product resin was washed with 4×1 mL N,N-dimethylformamide, 6×1 mL tetrahydrofuran, and 2×1 mL diethyl ether and then dried in vacuo to provide 26.2 mg free-flowing orange resin.

R_f 0.48 (methanolysis product, decomposes), 10% methanol–dichloromethane.

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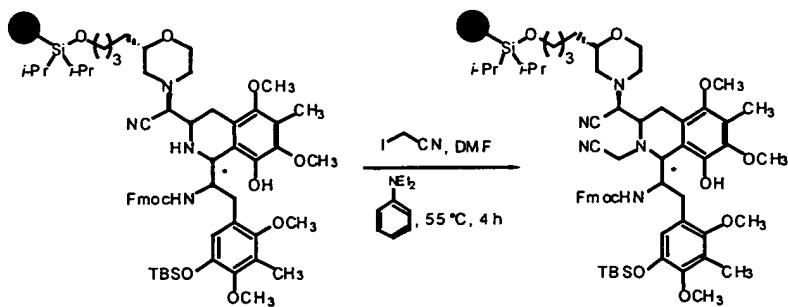
Zinc chloride (25.7 μ L, 0.5 M in tetrahydrofuran, 12.83 μ mol, 3.0 equiv) was added to a 20 mixture of boronate resin (26.2 mg, 4.28 μ mol, 0.1632 mmol/g, 1.0 equiv) and 4 Å molecular

sieves (2.5 mg) suspended in 160.0 μ L tetrahydrofuran. The resulting suspension was stirred at 55 °C for 1.5 h. The reaction supernatant was then removed via crimped cannula and collected along with 5 \times 1.5 mL dichloromethane washes of the product resin. The combined superntant and washes were partitioned between 15 mL dichloromethane and 15 mL aqueous phosphate buffer solution (0.05 M sodium phosphate monobasic, 0.05 M potassium phosphate dibasic, pH 7) and the organic layer isolated. The organic layer was then washed with an additional 8 mL phosphate buffer solution, dried over sodium sulfate, and concentrated in vacuo to provide an orangish-brown oil. Lyophilization of this oil from 300 μ L benzene afforded the heptacyclic product as a tan solid (2.2 mg, 57.7% yield (9 steps)).

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¹H NMR (600 MHz, CDCl₃), δ 7.53 (d, 2H, J = 7.8 Hz, ArH), 7.16 (d, 2H, J = 7.8 Hz, ArH), 5.58 (s, 1H, ArOH), 5.30 (s, 1H, ArOH), 4.89 (s, 1H, CHAr₁Ar₂), 4.23 (d, 1H, J = 3.0 Hz, N(CH₃)CH), 3.80 (s, 3H, ArOCH₃), 3.73 (s (obsc.), 1H, CHCN), 3.71 (s, 6H, 2 \times ArOCH₃), 3.64 (s, 2H, BOCH₂), 3.63 (s, 2H, BOCH₂), 3.47 (s, 3H, ArOCH₃), 3.39 (dt, 1H, J = 10.8 Hz, N(CH₃)CHCH), 3.31 (dd, 1H, J = 2.4, 16.2 Hz, N(CH₃)CHCHCH₂), 3.25 (d, 1H, J = 8.4 Hz, NCH(CN)CH), 2.83 (dd, 1H, J = 8.4, 18.0 Hz, NCH(CN)CHCH₂), 2.26 (s, 3H, NCH₃), 2.25 (s, 3H, ArCH₃), 2.18 (dd, 1H, J = 5.4, 15.6 Hz, N(CH₃)CHCHCH₂), 2.15 (s, 3H, ArCH₃), 1.84 (d, 1H, J = 18.0 Hz, NCH(CN)CHCH₂), 0.98 (s, 6H, C(CH₂)₂). R_f 0.27 (decomposes), 5% methanol-dichloromethane. HRMS (TOF-ES⁺) Calcd for C₃₈H₄₇BN₃O₈⁺ (M+H)⁺: 684.3456, 20 Found: 684.3487.

Exemplary alternate N-alkylation reaction:

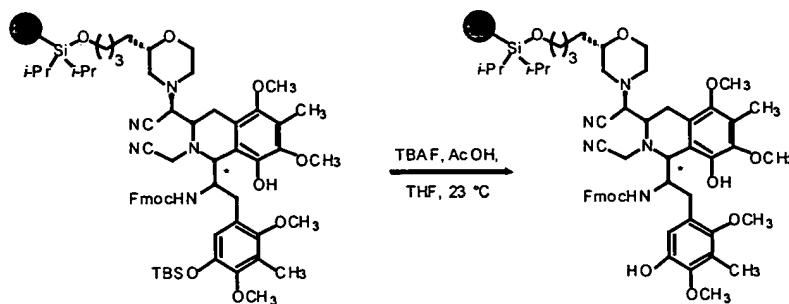


25

N,N-diethylaniline (34.9 μ L, 219.0 μ mol, 45.0 equiv) was added to a suspension of amine resin (23.5mg, 4.87 μ mol, 0.2071 mmol/g, 1.0 equiv) in 390.0 μ L N,N-diethylformamide. Iodoacetonitrile (14.1 μ L, 194.67 μ mol, 40.0 equiv) was then added and the resulting suspension stirred for 4 h at 55 °C. The reaction supernatant was then removed via a crimped cannula and 5 the product resin washed with 4 \times 2 mL N,N-dimethylformamide, 4 \times 2 mL tetrahydrofuran, and 2 \times 2 mL diethyl ether. The product resin was then dried in vacuo to provide a free-flowing red-brown resin.

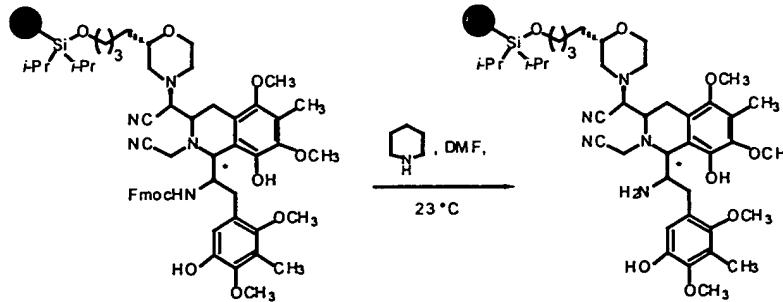
R_f 0.41, 0.45 (methanolysis products, diastereomers), 10% methanol–dichloromethane.

10

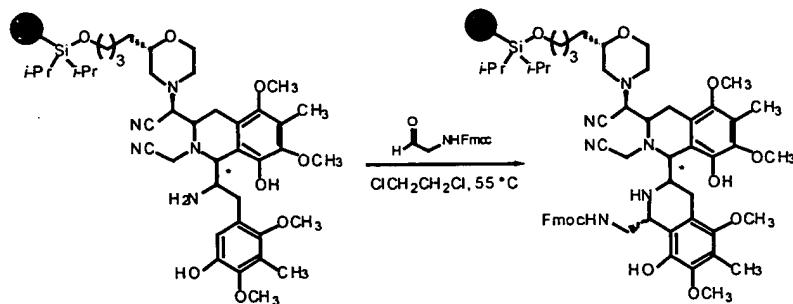


Acetic acid (1.1 μ L, 18.65 μ mol, 4.0 equiv) and tetrabutylammonium fluoride (1.0 M in tetrahydrofuran, 9.3 μ L, 9.30 μ mol, 2.0 equiv) were sequentially added to a suspension of 15 silylphenol resin (22.7 mg, 4.66 μ mol, 0.2054 mmol/g, 1.0 equiv) in 230 μ L tetrahydrofuran. The resulting suspension was stirred at 23 °C for 2.5 h before removal of the supernatant solution via crimped cannula. Washing of the product resin with 4 \times 1 mL N,N-dimethylformamide, 4 \times 1 mL tetrahydrofuran, and 2 \times 1 mL diethyl ether and drying in vacuo afforded the phenol resin as a free-flowing brown-orange resin.

20



Piperidine (60.0 μ L, 620.57 μ mol, 133 equiv) was added to a suspension of phenol resin (22.2 μ g, 4.66 μ mol, 1.0 equiv) in 240 μ L N,N-diethylformamide and the resulting suspension was stirred for 3.5 h at 23 °C. The supernatant solution was then removed via crimped cannula 5 and the product resin washed with 4×1 mL N,N-dimethylformamide, 6×1 mL tetrahydrofuran, and 2×1 mL diethyl ether. Drying of the washed resin in vacuo provided the immbolized aminophenol as a free-flowing orange resin.

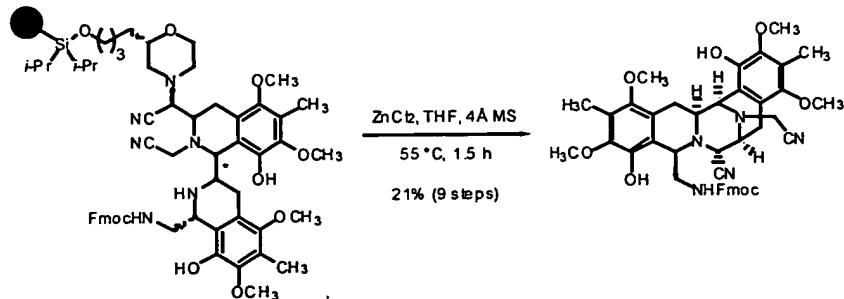


10

A solution of N-Fmoc glycinal (3.9 mg, 13.98 μ mol, 3.0 equiv) in 350 μ L 1,2-dichloroethane was freeze-pump-thaw deoxygenated (3 cycles) and then added via cannula to the aminophenol resin (21.2 mg, 4.66 μ mol, 1.0 equiv). The resulting suspension was stirred at 55 °C for 17 h before the supernatant solution was removed via crimped cannula. The product resin 15 was then washed with 3×1 mL N,N-dimethylformamide, 5×1 mL tetrahydrofuran, and 2×1 mL diethyl ether and dried in vacuo to afford the immbolized bistetrahydroisoquinoline as a free-flowing brown-orange resin.

R_f 0.47 (methanolysis product), 10% methanol–dichloromethane.

20



Zinc chloride (29.0 μ L, 0.5 M in tetrahydrofuran, 14.51 μ mol, 3.0 equiv) was added to a mixture of N-Fmoc resin (23.2 mg, 4.84 μ mol, 0.2085 mmol/g, 1.0 equiv) and 4 \AA molecular sieves (2.5 mg) suspended in 180.0 μ L tetrahydrofuran. The resulting suspension was stirred at 5 55 °C for 1.5 h before being diluted with 650 μ L dichloromethane. The resulting suspension was loaded onto a short silica gel plug and eluted with 4 plug-volumes of 10% tetrahydrofuran–dichloromethane. The eluted solvent was concentrated in vacuo to afford the product bis-aminonitrile as an off-white solid (0.8 mg, 21.4% yield (9 steps)).

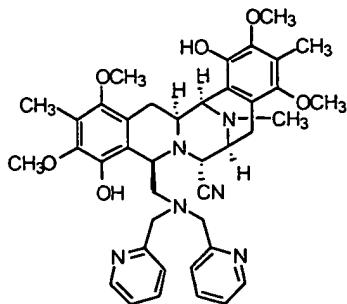
10 ^1H NMR (600 MHz, CDCl_3), δ 7.78 (m, 2H, J = 7.8 Hz, ArH), 7.60 (d, 1H, J = 7.8 Hz, ArH), 7.49 (m, 1H, ArH), 7.41 (m, 2H, ArH), 7.32 (m, 2H, ArH), 5.56 (s, 1H, ArOH), 5.54 (s, 1H, ArOH), 4.47 (t, 1H, NHFmoc), 4.31 (t, 1H, J = 6.0 Hz, CO_2CH_2), 4.29 (s, 1H, N(CH₂CN)CHAR), 4.24 (t, 1H, J = 7.8 Hz, CO_2CH_2), 4.11 (t, 1H, J = 4.2 Hz, NCHCH₂NHFmoc), 4.07 (t, 1H, $\text{CO}_2\text{CH}_2\text{CH}$), 3.75 (s, 3H, ArOCH₃), 3.73 (s, 3H, ArOCH₃), 3.63 (s, 1H, NCH(CN)CH), 15 3.61 (s, 3H, ArOCH₃), 3.48 (s, 3H, ArOCH₃), 3.40 (d, 1H, J = 16.8 Hz, NCH₂CN), 3.34 (d, 1H, J = 16.8 Hz, NCH₂CN), 3.28–3.17 (m, 3H, CH₂NHFmoc, N(CH₂CN)CHCH, N(CH₂CN)CHCHCH₂), 2.93–2.85 (m, 2H, CH₂NHFmoc, NCH(CN)CHCH₂), 2.36 (d, 1H, J = 19.2 Hz, NCH(CN)CHCH₂), 2.19 (s, 3H, ArCH₃), 2.17 (s, 3H, ArCH₃), 1.85 (m, 1H, N(CH₂CN)CHCHCH₂). FTIR (neat film), cm^{-1} 3350 (m, OH), 2925 (m), 1714 (s), 1455 (m), 20 1415 (m), 1109 (s), 1059 (s), 760 (m). R_f 0.50, 5% methanol–dichloromethane. HRMS (TOF-ES⁺) Calcd for C₄₄H₄₆N₅O₈⁺ (M+H)⁺: 772.3346, Found: 772.3369.

4) Synthesis and characterization of certain exemplary analogues:

It will be appreciated that although the synthesis of certain of the compounds are described using traditional solution phase techniques (for example as described in section 1 and 2 above) and the synthesis of certain of the compounds are described using solid-supported techniques (for example, as described in section 3 above), each of the compounds described below can be prepared using *either* traditional solution phase techniques or solid-supported techniques.

30

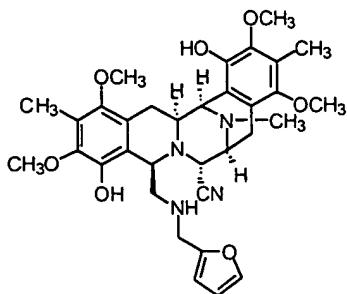
Example 1: bis-2-Pyridyl derivative



Pyridine 2-carboxaldehyde (0.29 μ L, 3.0 μ mol, 2.0 equiv) and sodium triacetoxyborohydride (0.48 mg, 2.3 μ mol, 1.5 equiv) were added sequentially, each in one portion, to a stirred solution of the amine (0.8 mg, 1.5 μ mol, 1 equiv) in acetonitrile (0.1 mL) at 24 °C under an argon atmosphere. The mixture was stirred for 1 h and then a second portion of pyridine 2-carboxaldehyde (0.29 μ L, 3.0 μ mol, 2.0 equiv) and sodium triacetoxyborohydride (0.48 mg, 2.3 μ mol, 1.5 equiv) were added sequentially, each in one portion. The mixture was stirred for a further 30 min, then was diluted with ethyl acetate (10 mL) and washed with a 1:1 mixture of brine solution and saturated aqueous sodium hydrogen carbonate solution (2×3 mL). The organic layer was dried over sodium sulfate and concentrated in vacuo to leave a colorless oil. Purification by flash column chromatography (ethyl acetate \rightarrow 80% ethyl acetate-methanol) gave the bis-2-pyridyl derivative (0.8 mg, 85%) as a white solid.

$^1\text{H NMR}$ (500 MHz, CDCl_3), δ 10.47 (br. s, 1H, NH), 8.51 (d, 1H, $J = 4.9$, ArH), 7.64 (ddd, 1H, $J = 9.4, 7.8, 1.7$, ArH), 7.37 (d, 1H, $J = 7.8$, ArH), 7.19 (m, 1H, ArH), 5.48 (s, 1H, ArOH), 4.27 (d, 1H, $J = 2.5$, $\text{CHC}\equiv\text{N}$), 4.10 (br. d, 1H, $J = \sim 1.5$, ArCHNCH₃), 4.00 (dd, 1H, $J = 9.3, 2.5$, ArCHCH₂NH), 3.82 (s, 3H, ArOCH₃), 3.81 (s, 4H, NCH₂Ar), 3.75 (s, 3H, ArOCH₃), 3.62 (s, 3H, ArOCH₃), 3.58 (s, 3H, ArOCH₃), 3.38 (br. d, 1H, $J = \sim 9.8$, CHCHC≡N), 3.19-3.12 (m, 2H, ArCHCHCH₂Ar, ArCHCHCH₂Ar), 3.05 (dd, 1H, $J = 18.1, 8.3$, $\text{CH}_2\text{CHCHC}\equiv\text{N}$), 2.88 (dd, 1H, $J = 13.7, 2.4$, ArCHCH₂N), 2.62 (dd, 1H, $J = 13.7, 9.3$, ArCHCH₂N), 2.45 (d, 1H, $J = 18.1$, $\text{CH}_2\text{CHCHC}\equiv\text{N}$), 2.28 (s, 3H, NCH₃), 2.23 (s, 3H, ArCH₃), 2.18 (s, 3H, ArCH₃), 1.73 (dd, 1H, $J = 14.9, 11.0$, ArCHCHCH₂). FTIR (neat film), cm^{-1} 3354, 2923, 2687, 2226, 1456. HRMS (ES) Calcd for $\text{C}_{40}\text{H}_{47}\text{N}_6\text{O}_6$ (MH^+): 707.3557, Found: 707.3580.

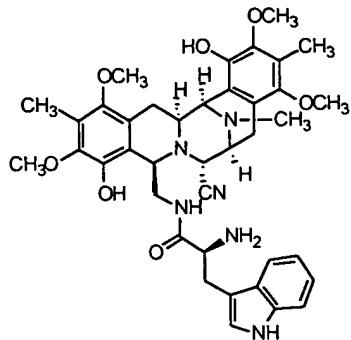
25 **Example 2: 2-Furylmethyl derivative**



Furaldehyde (0.32 μ L, 3.8 μ mol, 2.0 equiv) and sodium triacetoxyborohydride (0.61 mg, 2.9 μ mol, 1.5 equiv) were added sequentially, each in one portion, to a stirred solution of the 5 amine (1.0 mg, 1.9 μ mol, 1 equiv) in acetonitrile (0.1 mL) at 24 °C under an argon atmosphere. The mixture was stirred for 50 min, then was diluted with ethyl acetate (10 mL) and washed with a 1:1 mixture of brine solution and saturated aqueous sodium hydrogen carbonate solution (2 \times 3 mL). The organic layer was dried over sodium sulfate and concentrated in vacuo to leave a brown oil. Purification by flash column chromatography (80% ethyl acetate-hexanes) gave the 10 2-furylmethyl derivative (1.1 mg, 95%) as a white solid.

¹H NMR (500 MHz, CDCl₃), δ 7.34 (dd, 1H, *J* = 1.0, 0.9, ArH), 6.30 (dd, 1H, *J* = 3.2, 1.7, ArH), 6.10 (app. d, 1H, *J* = ~2.4, ArH), 5.52 (s, 1H, ArOH), 4.15 (dd, 1H, *J* = 2.9, 1.0, ArCHNCH₃), 4.00 (dd, 1H, *J* = 7.9, 3.0, ArCHCH₂NH), 3.98 (d, 1H, *J* = 2.5, CHC≡N), 3.79 (s, 3H, ArOCH₃), 3.76 (s, 3H, ArOCH₃), 3.72 (qu, 2H, AB system, ArCH₂NH), 3.61 (s, 3H, ArOCH₃), 3.60 (s, 3H, ArOCH₃), 3.38 (br. d, 1H, *J* = ~9.8, CHCHC≡N), 3.25-3.21 (m, 2H, ArCHCHCH₂Ar, ArCHCHCH₂Ar), 3.05 (dd, 1H, *J* = 18.5, 8.0, CH₂CHCHC≡N), 2.66 (dd, 1H, *J* = 13.2, 3.4, ArCHCH₂NH), 2.60 (dd, 1H, *J* = 13.2, 8.1, ArCHCH₂NH), 2.36 (d, 1H, *J* = 18.5, CH₂CHCHC≡N), 2.31 (s, 3H, NCH₃), 2.22 (s, 3H, ArCH₃), 2.19 (s, 3H, ArCH₃), 1.82 (dd, 1H, *J* = 15.6, 12.2, ArCHCHCH₂Ar). FTIR (neat film), cm⁻¹ 3395, 3313, 2933, 2226, 1462. HRMS 20 (ES) Calcd for C₃₃H₄₁N₄O₇ (MH)⁺: 605.2975, Found: 605.2956.

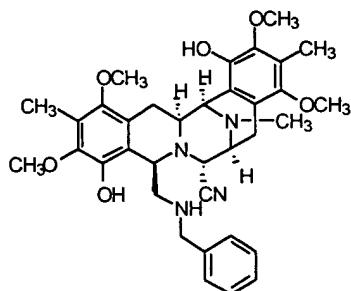
Example 3: L-Tryptophan derivative



Palladium on carbon (10 wt%, 0.18 mg, 0.17 μmol , 0.1 equiv) was added in one portion to the carbobenzyloxy-protected tryptophan derivative (1.5 mg, 1.7 μmol , 1 equiv) in methanol (0.5 mL) at 23 °C. A hydrogen atmosphere was introduced by alternatively evacuating the reaction flask and flushing with hydrogen. The reaction mixture was stirred for 3 h 20 min, then was filtered through a short pad of Celite. Concentration in vacuo left a white solid, which was purified by flash column chromatography (ethyl acetate \rightarrow 10:1 ethyl acetate-methanol) to give the *L*-tryptophan derivative (0.5 mg, 42%) as a white solid along with recovered starting material (0.5 mg, 33%).

¹H NMR (500 MHz, CDCl₃), δ 8.17 (br. s, 1H, ArNH), 7.56 (d, 1H, J = 7.8, ArH), 7.37 (d, 1H, J = 8.3, ArH), 7.20 (m, 1H, ArH), 7.11 (m, 1H, ArH), 6.84 (t, 1H, J = 5.6, NHCO), 6.69 (d, 1H, J = 2.4, ArH), 5.40 (s, 1H, ArOH), 5.31 (s, 1H, ArOH), 4.14-4.10 (m, 2H, CHCH₂NHCO, ArCHNCH₃), 3.85 (d, 1H, J = 2.4, CHC≡N), 3.76 (s, 3H, ArOCH₃), 3.70 (s, 3H, ArOCH₃), 3.61 (s, 3H, ArOCH₃), 3.66 (ddd, 1H, J = 13.6, 6.8, 3.3, CH₂NHCO), 3.49 (s, 3H, ArOCH₃), 3.34-3.27 (m, 1H (CHCHC≡N), 1H (CH₂NHCO), 1H (NHCOCHNH₂)), 3.23 (ddd, 1H, J = 11.7, 2.4, 2.4, ArCHCHCH₂Ar), 3.16 (dd, 1H, J = 9.0, 2.4, ArCHCHCH₂Ar), 3.02 (dd, 1H, J = 14.6, 4.8, NHCOCHCH₂), 2.95 (dd, 1H, J = 18.6, 7.9, CH₂CHCHC≡N), 2.69 (dd, 1H, J = 14.6, 7.3, NHCOCHCH₂), 2.41 (d, 1H, J = 18.6, CH₂CHCHC≡N), 2.28 (s, 3H, NCH₃), 2.21 (s, 3H, ArCH₃), 2.08 (s, 3H, ArCH₃), 1.89 (dd, 1H, J = 15.0, 10.7, ArCHCHCH₂). FTIR (neat film), cm⁻¹ 3354, 2913, 2226, 1651, 1262. HRMS (ES) Calcd for C₃₉H₄₆N₆O₇ (MH)⁺: 711.3506, Found: 711.3488.

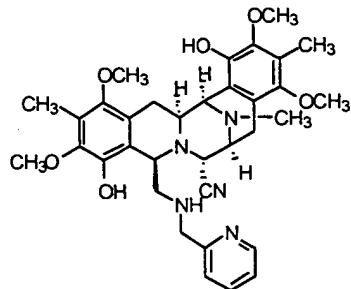
Example 4: Benzyl derivative



Benzaldehyde (0.43 μ L, 4.2 μ mol, 2.0 equiv) and sodium triacetoxyborohydride (0.67 mg, 3.1 μ mol, 1.5 equiv) were added sequentially, each in one portion, to a stirred solution of the amine (1.0 mg, 1.9 μ mol, 1 equiv) in acetonitrile (0.1 mL) at 23 °C under an argon atmosphere. The mixture was stirred for 45 min, then was diluted with ethyl acetate (10 mL) and washed with a 1:1 mixture of brine solution and saturated aqueous sodium hydrogen carbonate solution (2 \times 3 mL). The organic layer was dried over sodium sulfate and concentrated in vacuo to leave a colorless oil. Purification by flash column chromatography (ethyl acetate) gave the benzyl derivative (1.2 mg, 93%) as a white solid.

^1H NMR (500 MHz, CDCl_3), δ 7.33-7.28 (m, 3H, ArH), 7.19-7.17 (m, 2H, ArH), 5.52 (s, 1H, ArOH), 5.31 (s, 1H, ArOH), 4.14 (dd, 1H, J = 2.9, 1.0, ArCHNCH₃), 4.01 (t, 1H, J = 5.6, ArCHCH₂NH), 3.86 (d, 1H, J = 2.4, CHC≡N), 3.80 (s, 3H, ArOCH₃), 3.75 (s, 3H, ArOCH₃), 3.73 (d, 2H, J = 3.0, ArCH₂NH), 3.61 (s, 3H, ArOCH₃), 3.56 (s, 3H, ArOCH₃), 3.35 (br. d, 1H, J = ~7.8, CHCHC≡N), 3.25-3.21 (m, 2H, ArCHCHCH₂Ar, ArCHCHCH₂Ar), 3.02 (dd, 1H, J = 18.6, 7.8, CH₂CHCHC≡N), 2.66 (d, 1H, J = 5.3, ArCHCH₂NH), 2.33 (d, 1H, J = 18.6, CH₂CHCHC≡N), 2.30 (s, 3H, NCH₃), 2.20 (s, 6H, 2 ∞ ArCH₃), 1.83 (dd, 1H, J = 15.8, 12.0, ArCHCHCH₂Ar). **FTIR** (neat film), cm^{-1} 3395, 2923, 2574, 2226, 1456, 1108. **HRMS (ES)** Calcd for C₃₅H₄₃N₄O₆ (MH)⁺: 615.3182, Found: 615.3176.

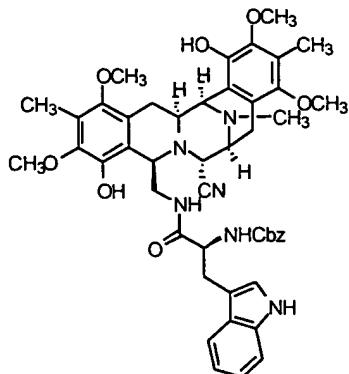
Example 5: 2-Pyridyl derivative



Pyridine 2-carboxaldehyde (0.36 μ L, 3.8 μ mol, 2.0 equiv) and sodium triacetoxyborohydride (0.61 mg, 2.9 μ mol, 1.5 equiv) were added sequentially, each in one portion, to a stirred solution of the amine (1.0 mg, 1.9 μ mol, 1 equiv) in acetonitrile (0.15 mL) at 24 °C under an argon atmosphere. The mixture was stirred for 40 min, then was diluted with ethyl acetate (10 mL) and washed with a 1:1 mixture of brine solution and saturated aqueous sodium hydrogen carbonate solution (2×3 mL). The organic layer was dried over sodium sulfate and concentrated in vacuo to leave a white solid. Purification by flash column chromatography (ethyl acetate \rightarrow 90% ethyl acetate-methanol) gave the 2-pyridyl derivative (0.6 mg, 51%) as a white solid.

$^1\text{H NMR}$ (500 MHz, CDCl_3), δ 8.53-8.52 (m, 1H, ArH), 7.63 (ddd, 1H, $J = 7.8, 7.8, 2.0$, ArH), 7.19 (ddd, 1H, $J = 7.8, 5.8, 1.0$, ArH), 7.14 (d, 1H, $J = 7.8$, ArH), 5.51 (s, 1H, ArOH), 4.15 (dd, 1H, $J = 2.5, 1.0$, ArCHNCH₃), 4.07 (d, 1H, $J = 2.4$, CHC≡N), 4.05 (dd, 1H, $J = 8.3, 2.5$, ArCHCH₂NH), 3.86 (q, 2H, AB system, ArCH₂NH), 3.78 (s, 3H, ArOCH₃), 3.75 (s, 3H, ArOCH₃), 3.61 (s, 3H, ArOCH₃), 3.52 (s, 3H, ArOCH₃), 3.39 (br. d, 1H, $J = \sim 8.8$, CHCHC≡N), 3.26-3.21 (m, 2H, ArCHCHCH₂Ar, ArCHCHCH₂Ar), 3.03 (dd, 1H, $J = 18.1, 7.6$, CH₂CHCHC≡N), 2.75 (dd, 1H, $J = 12.7, 2.6$, ArCHCH₂NH), 2.62 (dd, 1H, $J = 12.7, 8.5$, ArCHCH₂NH), 2.35 (d, 1H, $J = 18.1$, CH₂CHCHC≡N), 2.31 (s, 3H, NCH₃), 2.19 (s, 3H, ArCH₃), 2.19 (s, 3H, ArCH₃), 1.83 (dd, 1H, $J = 15.6, 12.2$, ArCHCHCH₂Ar). **FTIR** (neat film), cm^{-1} 3303, 2923, 2226, 1456, 1108. **HRMS (ES)** Calcd for C₃₄H₄₂N₅O₆ (MH)⁺: 616.3135, Found: 616.3156.

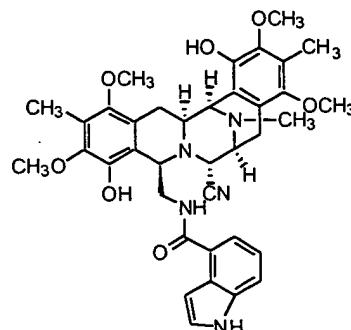
Example 6: N-Carbobenzyloxy-L-tryptophan derivative



Diethylaniline (0.60 μ L, 3.8 μ mol, 1.1 equiv) was added in one portion to a stirred solution of the amine (1.8 mg, 3.4 μ mol, 1 equiv) in THF (0.3 mL) at 0 °C under an argon atmosphere and the solution was stirred for 5 min. *N*-Carbobenzyloxy-*L*-tryptophan (1.5 mg, 4.5 μ mol, 1.3 equiv), 1-[3-(dimethylamino)propyl]-3-ethylcarbodiimide hydro-chloride (0.85 mg, 4.5 μ mol, 1.3 equiv) and 1-hydroxybenzotriazole (0.56 mg, 4.1 μ mol, 1.2 equiv) were then added separately, each in one portion, to the above solution at 0 °C. The reaction mixture was warmed to 23 °C over 13 h 55 min, then was quenched with saturated aqueous ammonium chloride solution (3 mL). The mixture was diluted with ethyl acetate (10 mL) and the layers were separated. The aqueous layer was extracted with ethyl acetate (10 mL) and the combined organic layer was dried over sodium sulfate. Concentration in vacuo left a white solid, which was purified by flash column chromatography (65% ethyl acetate-hexanes) to give the *N*-carbobenzyloxy-*L*-tryptophan derivative (2.6 mg, 90%) as a white solid.

^1H NMR (500 MHz, CDCl_3), δ 8.20 (br. s, 1H, IndNH), 7.73 (d, 1H, J = 7.8, ArH), 7.41 (d, 1H, J = 7.8, ArH), 7.35 (br. s, 3H, ArH), 7.23 (m, 2H, ArH), 7.17 (t, 1H, J = 7.3, ArH), 7.02 (d, 1H, J = 2.4, ArH), 5.71 (br. s, 1H, NHCO), 5.58 (s, 1H, ArOH), 5.51 (s, 1H, ArOH), 5.42 (d, 1H, J = 7.8, CbzNH), 5.11 (s, 2H, ArCH₂O), 4.36-4.32 (br. m, 1H), 4.04 (d, 1H, J = 1.5, CHC≡N), 3.84-3.81 (br. m, 1H), 3.74 (s, 3H, ArOCH₃), 3.70 (s, 6H, 2xArOCH₃), 3.57 (s, 3H, ArOCH₃), 3.24-3.21 (br. m, 2H), 3.16 (dd, 1H, J = 15.6, 2.4, ArCHCHCH₂Ar), 3.09-3.04 (m, 3H), 2.83-2.80 (br. m, 1H), 2.68-2.61 (br. m, 2H), 2.24 (s, 3H, NCH₃), 2.19 (s, 3H, ArCH₃), 2.17 (s, 3H, ArCH₃), 1.97 (d, 1H, J = 18.6, CH₂CHCHC≡N), 1.74 (dd, 1H, J = 15.6, 12.2, ArCHCHCH₂). **FTIR** (neat film), cm^{-1} 3344, 2923, 1708, 1672, 1456. **HRMS (ES)** Calcd for $\text{C}_{47}\text{H}_{53}\text{N}_6\text{O}_9$ (MH^+): 845.3874, Found: 845.3910.

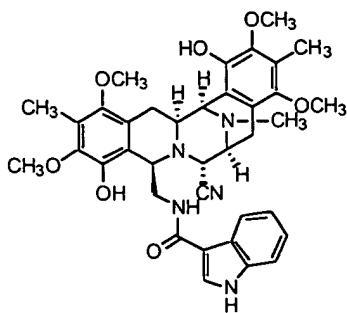
Example 7: Indole-4-carboxylic acid amide derivative



Indole-4-carboxylic acid (0.51 mg, 3.2 μ mol, 1.5 equiv), was added in one portion to a stirred solution of the amine (1.1 mg, 2.1 μ mol, 1 equiv) in dichloromethane (0.15 mL) at 23 °C under an argon atmosphere and the solution was stirred for 10 min. *N*-Cyclohexylcarbodiimide-*N'*-propyloxymethyl polystyrene (Argonaut Technologies, 1.13 mmol/g, 3.7 mg, 4.2 μ mol, 2.0 equiv) was then added, in one portion, to the above solution at 23 °C. The reaction mixture was stirred gently at 23 °C for 18 h 20 min, then was purified by flash column chromatography (80% ethyl acetate-hexanes) to give the indole-4-carboxylic acid amide derivative (0.8 mg, 60%) as a white solid.

$^1\text{H NMR}$ (500 MHz, CDCl_3), δ 8.19 (s, 1H, ArNH), 7.40 (d, 1H, $J = 7.8$, ArH), 7.16 (t, 1H, $J = 3.0$, ArH), 6.97 (t, 1H, $J = 7.8$, ArH), 6.69 (bt, 1H, $J = \sim 2.0$, ArH), 6.52 (d, 1H, $J = 7.3$, ArH), 6.00 (d, 1H, $J = 6.3$, NH), 5.71 (s, 1H, ArOH), 5.59 (s, 1H, ArOH), 4.34 (br. s, 1H, CHCH_2NHCO), 4.28 (ddd, 1H, $J = 13.7$, 8.3, 2.0, CH_2NHCO), 4.22 (bd, 1H, $J = \sim 2.0$, ArCHNCH₃), 4.15 (d, 1H, $J = 2.9$, $\text{CHC}\equiv\text{N}$), 3.75 (s, 3H, ArOCH₃), 3.74 (s, 3H, ArOCH₃), 3.45 (br. d, 1H, $J = \sim 7.3$, $\text{CHCHC}\equiv\text{N}$), 3.40 (s, 3H, ArOCH₃), 3.38-3.33 (m, 1H (CH_2NHCO), 1H (ArCHCHCH₂Ar)), 3.29 (s, 3H, ArOCH₃), 3.28 (app. dd (obsc.), 1H, $J = \sim 16.1$, 2.9, ArCHCHCH₂Ar), 3.08 (dd, 1H, $J = 18.6$, 8.0, $\text{CH}_2\text{CHCHC}\equiv\text{N}$), 2.43 (d, 1H, $J = 18.6$, $\text{CH}_2\text{CHCHC}\equiv\text{N}$), 2.31 (s, 3H, NCH₃), 2.15 (s, 3H, ArCH₃), 2.08 (dd, 1H, $J = 16.1$, 11.7, ArCHCHCH₂Ar), 2.01 (s, 3H, ArCH₃). **FTIR** (neat film), cm^{-1} 3393, 2926, 2227, 1641. **HRMS (ES)** Calcd for $\text{C}_{37}\text{H}_{41}\text{N}_5\text{O}_7$ (MH^+): 668.3084, Found: 668.3062.

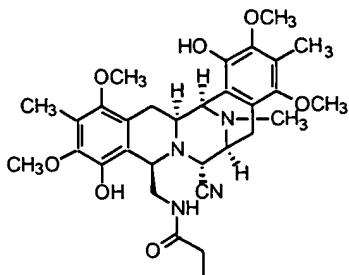
Example 8: Indole-3-carboxylic acid amide derivative



Indole-3-carboxylic acid (0.55 mg, 3.4 μ mol, 1.5 equiv), was added in one portion to a
5 stirred solution of the amine (1.2 mg, 2.2 μ mol, 1 equiv) in dichloromethane (0.2 mL) at 23 °C
under an argon atmosphere and the solution was stirred for 10 min. *N*-Cyclohexylcarbodiimide-
N'-propyloxymethyl polystyrene (Argonaut Technologies, 1.13 mmol/g, 8.0 mg, 9.2 μ mol, 4.0
equiv) was then added, in one portion, to the above solution at 23 °C. The reaction mixture was
stirred gently at 23 °C for 16 h 45 min, then was purified by flash column chromatography (85%
10 ethyl acetate-hexanes) to give the indole-3-carboxylic acid amide derivative (1.2 mg, 86%) as a
white solid.

¹H NMR (500 MHz, CDCl₃), δ 8.20 (br. s, 1H, ArNH), 7.80 (d, 1H, J = 8.3, ArH), 7.32
(app. d, 1H, J = ~7.4, ArH), 7.18 (td, 1H, J = 7.4, 1.3, ArH), 7.08 (td, 1H, J = 7.4, 1.0, ArH),
6.55 (d, 1H, J = 2.9, ArH), 5.75 (s, 1H, ArOH), 5.67 (br. d, 1H, J = ~5.9, NH), 5.61 (s, 1H,
15 ArOH), 4.31 (br. s, 1H, CHCH₂NHCO), 4.23 (dd, 1H, J = 3.0, 1.0, ArCHNCH₃), 4.16-4.12 (m,
2H, CHC≡N, CH₂NHCO), 3.81 (s, 3H, ArOCH₃), 3.74 (s, 3H, ArOCH₃), 3.45 (br. s, 4H,
ArOCH₃, CHCHC≡N), 3.37 (ddd, 1H, J = 12.0, 2.6, 2.6, ArCHCHCH₂Ar), 3.34 (s, 3H,
ArOCH₃), 3.30 (ddd, 1H, J = 13.7, 3.9, 2.7, CH₂NHCO), 3.26 (dd, 1H, J = 15.8, 2.6,
20 ArCHCHCH₂Ar), 3.10 (dd, 1H, J = 18.5, 7.8, CH₂CHCHC≡N), 2.49 (d, 1H, J = 18.5,
CH₂CHCHC≡N), 2.34 (s, 3H, NCH₃), 2.14 (s, 3H, ArCH₃), 2.06 (s, 3H, ArCH₃), 2.02 (dd, 1H, J
= 15.8, 11.7, ArCHCHCH₂Ar). **FTIR** (neat film), cm⁻¹ 3388, 2925, 2225, 1620. **HRMS (ES)**
Calcd for C₃₇H₄₁N₅O₇ (MH)⁺: 668.3084, Found: 668.3062.

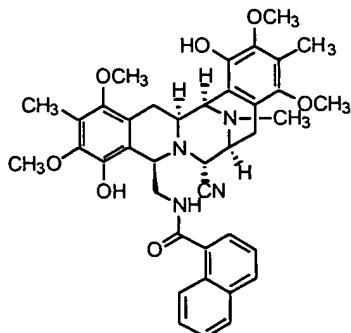
Example 9: Propionamide derivative



Diethylaniline (0.60 μ L, 4.0 μ mol, 1.1 equiv) and propionyl chloride (1.0 mg, 1.0 μ L, 5 μ mol, 3.0 equiv) were added separately, each in one portion, to a stirred solution of the amine (1.9 mg, 3.6 μ mol, 1 equiv) in dichloromethane (0.2 mL) at 0 °C under an argon atmosphere. The reaction mixture was stirred at 0 °C for 30 min, then was quenched by the addition of a 1:1 mixture of water and saturated aqueous sodium hydrogen carbonate solution (4 mL). The mixture was extracted with ethyl acetate (2 \times 15 mL) and the combined organic layer 10 was washed with a 1:1 mixture of brine solution and saturated aqueous sodium hydrogen carbonate solution (5 mL). The organic layer was dried over sodium sulfate and concentrated in vacuo to leave a colorless oil. Purification by flash column chromatography (ethyl acetate \rightarrow 90% ethyl acetate-methanol) gave the propionamide derivative (1.9 mg, 90%) as a white solid.

¹H NMR (500 MHz, CDCl₃), δ 5.63 (s, 1H, ArOH), 5.60 (s, 1H, ArOH), 5.21 (d, 1H, J = 5.9, NH), 4.20 (d, 1H, J = 2.0, ArCHNCH₃), 4.17 (bd, 1H, J = 2.9, CHCH₂NHCO), 4.03 (d, 1H, J = 2.4, CHC≡N), 3.84 (ddd, 1H, J = 13.4, 8.3, 1.9, CH₂NHCO), 3.78 (s, 3H, ArOCH₃), 3.75 (s, 3H, ArOCH₃), 3.71 (s, 3H, ArOCH₃), 3.60 (s, 3H, ArOCH₃), 3.43 (br. d, 1H, J = ~7.8, CHCHC≡N), 3.32 (ddd, 1H, J = 11.9, 2.7, 2.7, ArCHCHCH₂Ar), 3.24 (dd, 1H, J = 15.8, 2.7, ArCHCHCH₂Ar), 3.12-3.07 (m, 2H, CH₂CHCHC≡N, CH₂NHCO), 2.50 (d, 1H, J = 18.5, 20 CH₂CHCHC≡N), 2.35 (s, 3H, NCH₃), 2.25 (s, 3H, ArCH₃), 2.19 (s, 3H, ArCH₃), 1.90 (dd, 1H, J = 15.8, 12.0, ArCHCHCH₂Ar), 1.56 (q, 2H, J = 7.8, CH₃CH₂CO), 0.74 (t, 3H, J = 7.8, CH₃CH₂CO). **FTIR** (neat film), cm⁻¹ 3385, 2923, 2226, 1657. **HRMS (ES)** Calcd for C₃₁H₄₀N₄O₇ (MH)⁺: 581.2975, Found: 581.2948.

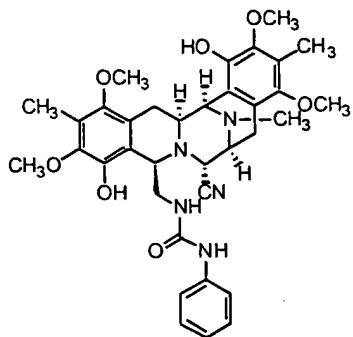
25 **Example 10: Naphthalene-1-carboxylic acid amide derivative**



Diethylaniline (0.27 μ L, 1.7 μ mol, 1.1 equiv) was added in one portion to a stirred solution of the amine (0.8 mg, 1.5 μ mol, 1 equiv) in THF (0.2 mL) at 0 °C under an argon atmosphere and the solution was stirred for 5 min. 1-Naphthoic acid (0.34 mg, 2.0 μ mol, 1.3 equiv), 1-[3-(dimethylamino)propyl]-3-ethylcarbodiimide hydrochloride (0.38 mg, 2.0 μ mol, 1.3 equiv) and 1-hydroxybenzotriazole (0.25 mg, 1.8 μ mol, 1.2 equiv) were then added separately, each in one portion, to the above solution at 0 °C. The reaction mixture was warmed to 23 °C over 14 h 45 min, then was quenched with saturated aqueous ammonium chloride solution (3 mL). The mixture was diluted with ethyl acetate (10 mL) and the layers were separated. The aqueous layer was extracted with ethyl acetate (10 mL) and the combined organic layer was dried over sodium sulfate. Concentration in vacuo left a white solid, which was purified by flash column chromatography (80% ethyl acetate-hexanes) to give the naphthalene-1-carboxylic acid amide derivative (0.7 mg, 68%) as a white solid.

¹H NMR (500 MHz, CDCl₃), δ 7.79-7.75 (m, 3H, ArH), 7.45-7.42 (m, 1H, ArH), 7.36 (ddd, 1H, J = 7.3, 6.8, 1.2, ArH), 7.19 (dd, 1H, J = 7.8, 6.8, ArH), 6.76 (app. d, 1H, J = ~6.8, ArH), 5.71 (s, 1H, ArOH), 5.67 (d, 1H, J = 4.9, NH), 5.49 (s, 1H, ArOH), 4.37-4.33 (m, 2H, CH₂NHCO, CHCH₂NHCO), 4.17 (d, 1H, J = 1.4, ArCHNCH₃), 4.16 (d, 1H, J = 2.9, CHC≡N), 3.77 (s, 3H, ArOCH₃), 3.55 (s, 3H, ArOCH₃), 3.45-3.39 (m, 5H, CHCHC≡N, ArOCH₃, CH₂NHCO), 3.34 (ddd, 1H, J = 12.0, 2.7, 2.7, ArCHCHCH₂Ar), 3.28 (s, 3H, ArOCH₃), 3.24 (dd, 1H, J = 16.1, 2.5, ArCHCHCH₂Ar), 3.05 (dd, 1H, J = 18.6, 8.0, CH₂CHCHC≡N), 2.43 (d, 1H, J = 18.6, CH₂CHCHC≡N), 2.29 (s, 3H, NCH₃), 2.20 (s, 3H, ArCH₃), 1.95 (dd, 1H, J = 16.1, 11.7, ArCHCHCH₂Ar), 1.88 (s, 3H, ArCH₃). FTIR (neat film), cm⁻¹ 3395, 2923, 2226, 1651. HRMS (ES) Calcd for C₃₉H₄₂N₄O₇ (MH)⁺: 679.3132, Found: 679.3106.

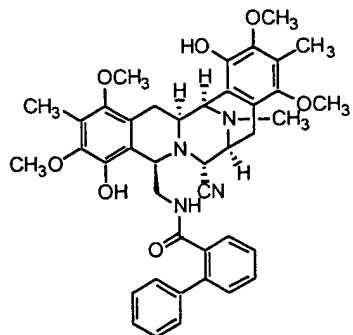
Example 11: Phenylurea derivative



5 Diethylaniline (0.33 μ L, 2.1 μ mol, 1.1 equiv) and phenyl isocyanate (0.25 μ L, 2.3 μ mol, 1.2 equiv) were added separately, each in one portion, to a stirred solution of the amine (1.0 mg, 1.9 μ mol, 1 equiv) in dichloromethane (0.2 mL) at 0 °C under an argon atmosphere. The reaction mixture was stirred at 0 °C for 25 min, then was quenched by the addition of a 1:1 mixture of water and saturated aqueous sodium hydrogen carbonate solution (4 mL). The
10 mixture was extracted with dichloromethane (2×10 mL) and the organic layer was dried over sodium sulfate and concentrated in vacuo to leave a white solid. Purification by flash column chromatography (70% ethyl acetate) gave the phenylurea derivative (0.9 mg, 74%) as a white solid.

15 **^1H NMR** (500 MHz, CDCl_3), δ 7.27-7.23 (m, 4H, ArH), 7.01-6.98 (m, 1H, ArH), 6.26 (s, 1H, NH), 5.66 (s, 1H, ArOH), 5.53 (s, 1H, ArOH), 4.36 (t, 1H, $J = 5.8$, NH), 4.23-4.21 (m, 2H, CHCH_2NHCO , ArCHNCH_3), 4.03 (d, 1H, $J = 2.4$, $\text{CHC}\equiv\text{N}$), 3.81 (s, 6H, $2 \times \text{ArOCH}_3$), 3.74 (s, 3H, ArOCH_3), 3.59 (s, 3H, ArOCH_3), 3.44 (ddd, 1H, $J = 13.7, 5.4, 5.4$, CH_2NHCO), 3.38 (br. d, 1H, $J = 7.3$, $\text{CHCHC}\equiv\text{N}$), 3.31 (ddd, 1H, $J = 12.7, 2.4, 2.4$, $\text{ArCHCHCH}_2\text{Ar}$), 3.21 (dd, 1H, $J = 15.6, 2.9$, $\text{ArCHCHCH}_2\text{Ar}$), 3.10 (dd, 1H, $J = 18.5, 7.3$, $\text{CH}_2\text{CHCHC}\equiv\text{N}$), 2.75-2.69 (m, 2H, CH_2NHCO , $\text{CH}_2\text{CHCHC}\equiv\text{N}$), 2.43 (s, 3H, NCH_3), 2.26 (s, 3H, ArCH_3), 2.19 (s, 3H, ArCH_3), 1.82 (dd, 1H, $J = 15.6, 12.5$, $\text{ArCHCHCH}_2\text{Ar}$). **FTIR** (neat film), cm^{-1} 3374, 2923, 2226, 1672. **HRMS (ES)** Calcd for $\text{C}_{35}\text{H}_{41}\text{N}_5\text{O}_7$ (MH^+): 644.3084, Found: 644.3110.

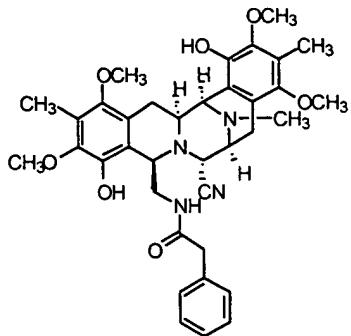
Example 12: Biphenyl-2-carboxylic acid amide derivative



5 2-Biphenylcarboxylic acid (0.51 mg, 2.6 μ mol, 1.5 equiv) was added in one portion to a stirred solution of the amine (0.9 mg, 1.7 μ mol, 1 equiv) in dichloromethane (0.2 mL) at 23 °C under an argon atmosphere and the solution was stirred for 10 min. *N*-Cyclohexylcarbodiimide-*N'*-propyloxymethyl polystyrene (Argonaut Technologies, 1.13 mmol/g, 3.0 mg, 3.4 μ mol, 2.0 equiv) was then added, in one portion, to the above solution at 23 °C. The reaction mixture was 10 stirred gently at 23 °C for 19 h 30 min, then was purified by flash column chromatography (70% ethyl acetate-hexanes) to give the biphenyl-2-carboxylic acid amide derivative (0.9 mg, 75%) as a white solid.

15 **^1H NMR** (500 MHz, CDCl_3), δ 7.40 (td, 1H, J = 7.3, 1.4, ArH), 7.33-7.29 (m, 4H, ArH), 7.24-7.22 (m, 2H, ArH), 7.20 (td, 1H, J = 7.3, 1.4, ArH), 6.96 (dd, 1H, J = 5.8, 1.0, ArH), 5.55 (s, 1H, ArOH), 5.53 (s, 1H, ArOH), 5.52 (t, 1H, J = 5.8, NH), 4.13 (d, 1H, J = 2.5, $\text{CHC}\equiv\text{N}$), 4.03 (t, 1H, J = 3.9, CHCH_2NHCO), 3.77 (d, 1H, J = 2.5, ArCHNCH_3), 3.73 (s, 3H, ArOCH₃), 3.72 (s, 3H, ArOCH₃), 3.58 (s, 3H, ArOCH₃), 3.45 (s, 3H, ArOCH₃), 3.45-3.40 (m, 1H, CH₂NHCO), 3.36-3.31 (m, 2H, CH₂NHCO, CHCHC≡N), 3.22-3.18 (m, 2H, ArCHCHCH₂Ar, ArCHCHCH₂Ar), 3.03-2.98 (dd, 1H, J = 18.6, 8.3, CH₂CHCHC≡N), 2.39 (d, 1H, J = 18.6, 20 CH₂CHCHC≡N), 2.27 (s, 3H, NCH₃), 2.20 (s, 3H, ArCH₃), 2.12 (s, 3H, ArCH₃), 1.86 (dd, 1H, J = 16.2, 12.2, ArCHCHCH₂Ar). **FTIR** (neat film), cm^{-1} 3402, 2930, 2231, 1654. **HRMS (ES)** Calcd for C₄₁H₄₄N₄O₇ (MH)⁺: 705.3288, Found: 705.3317.

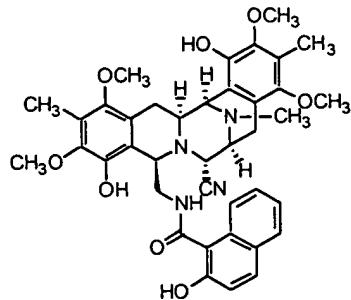
Example 13: Phenyl acetyl derivative



Diethylaniline (0.50 μ L, 2.9 μ mol, 1.1 equiv) and phenyl acetyl chloride (1.1 μ L, 8.0 μ mol, 3.0 equiv) were added separately, each in one portion, to a stirred solution of the amine 5 (1.4 mg, 2.7 μ mol, 1 equiv) in dichloromethane (0.15 mL) at 0 °C under an argon atmosphere. The reaction mixture was stirred at 0 °C for 30 min, then was quenched by the addition of a 1:1 mixture of water and saturated aqueous sodium hydrogen carbonate solution (4 mL). The mixture was extracted with ethyl acetate (2×15 mL) and the combined organic layer was washed with a 1:1 mixture of brine solution and saturated aqueous sodium hydrogen carbonate 10 solution (5 mL). The organic layer was dried over sodium sulfate and concentrated in vacuo to leave a light yellow oil. Purification by flash column chromatography (80% ethyl acetate-hexanes) gave the phenyl acetyl derivative (1.7 mg, 99%) as a white solid.

¹H NMR (500 MHz, CDCl₃), δ 7.21-7.17 (m, 3H, ArH), 6.90-6.89 (m, 2H, ArH), 5.59 (s, 1H, ArOH), 5.41 (t, 1H, J = 5.1, NHCO), 5.39 (s, 1H, ArOH), 4.17 (d, 1H, J = 1.9, 15 ArCHNCH₃), 4.09 (dd, 1H, J = 5.4, 2.4, CHCH₂NHCO), 4.02 (d, 1H, J = 2.5, CHC≡N), 3.75 (s, 3H, ArOCH₃), 3.74 (s, 3H, ArOCH₃), 3.71 (s, 3H, ArOCH₃), 3.66 (app. ddd (obsc), 1H, CH₂NHCO), 3.56 (s, 3H, ArOCH₃), 3.39 (br. d, 1H, J = ~8.8, CHCHC≡N), 3.23 (ddd, 1H, J = 11.7, 2.4, 2.4, ArCHCHCH₂Ar), 3.20-3.15 (m, 2H, CH₂NHCO, ArCHCHCH₂Ar)), 3.15 (d, 1H, J = 15.1, NHCOCH₂Ar), 3.06 (dd, 1H, J = 18.6, 8.3, CH₂CHCHC≡N), 2.96 (d, 1H, J = 15.1, 20 NHCOCH₂Ar), 2.51 (d, 1H, J = 18.6, CH₂CHCHC≡N), 2.32 (s, 3H, NCH₃), 2.24 (s, 3H, ArCH₃), 2.18 (s, 3H, ArCH₃), 1.81 (dd, 1H, J = 16.4, 12.0, ArCHCHCH₂Ar). FTIR (neat film), cm⁻¹ 3385, 2923, 2226, 1667, 1462. HRMS (ES) Calcd for C₃₆H₄₃N₄O₇ (MH)⁺: 643.3132, Found: 643.3130.

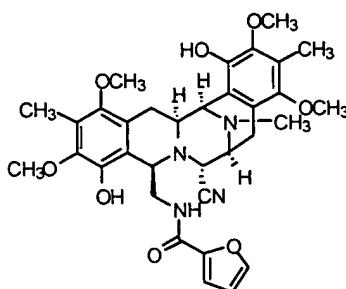
25 Example 14: 2-Hydroxy-naphthalene-1-carboxylic acid amide derivative



Diethylaniline (0.27 μ L, 1.7 μ mol, 1.1 equiv) was added in one portion to a stirred solution of the amine (0.8 mg, 1.5 μ mol, 1 equiv) in THF (0.2 mL) at 0 °C under an argon atmosphere and the solution was stirred for 5 min. 2-Hydroxy-1-naphthoic acid (0.37 mg, 2.0 μ mol, 1.3 equiv), 1-[3-(dimethylamino)propyl]-3-ethylcarbodiimide hydrochloride (0.38 mg, 2.0 μ mol, 1.3 equiv) and 1-hydroxybenzotriazole (0.25 mg, 1.8 μ mol, 1.2 equiv) were then added separately, each in one portion, to the above solution at 0 °C. The reaction mixture was warmed to 23 °C over 13 h 20 min, then was quenched with saturated aqueous ammonium chloride solution (3 mL). The mixture was diluted with ethyl acetate (10 mL) and the layers were separated. The aqueous layer was extracted with ethyl acetate (10 mL) and the combined organic layer was dried over sodium sulfate. Concentration in vacuo left a white solid, which was purified by flash column chromatography (85% ethyl acetate-hexanes) to give the 2-hydroxy-naphthalene-1-carboxylic acid amide derivative (1.1 mg, 100%) as a white solid.

1H NMR (500 MHz, CDCl₃), δ 11.22 (s, 1H, NaphOH), 7.70 (d, 1H, *J* = 8.8, ArH), 7.66 (d, 1H, *J* = 7.8, ArH), 7.28 (d (obsc.), 1H, ArH), 7.23 (t, 1H, *J* = 7.8, ArH), 7.11 (t, 1H, *J* = 7.8, ArH), 7.04 (d, 1H, *J* = 8.8, ArH), 6.10 (br. s, 1H, NH), 5.71 (s, 1H, ArOH), 5.46 (s, 1H, ArOH), 4.40 (br. s, 1H, CHCH₂NHCO), 4.27 (app. dd, 1H, *J* = ~13.8, 6.1, CH₂NHCO), 4.17-4.15 (m, 2H, ArCHNCH₃, CHC≡N), 3.75 (s, 3H, ArOCH₃), 3.68 (s, 3H, ArOCH₃), 3.54 (ddd, 1H, *J* = 14.2, 3.9, 3.9, ArCHCHCH₂Ar), 3.48 (s, 3H, ArOCH₃), 3.42 (br. d, 1H, *J* = ~7.9, CHCHC≡N), 3.38-3.33 (m, 1H, CH₂NHCO), 3.28 (dd, 1H, *J* = 16.2, 2.8, ArCHCHCH₂Ar), 3.15 (s, 3H, ArOCH₃), 2.99 (dd, 1H, *J* = 18.1, 8.3, CH₂CHCHC≡N), 2.32 (d, 1H, *J* = 18.1, CH₂CHCHC≡N), 2.25 (s, 3H, NCH₃), 2.24 (s, 3H, ArCH₃), 2.12 (dd, 1H, *J* = 16.2, 11.8, ArCHCHCH₂Ar), 1.78 (s, 3H, ArCH₃). **FTIR** (neat film), cm⁻¹ 3395, 2923, 2226, 1626. **HRMS (ES)** Calcd for C₃₉H₄₂N₄O₈ (MH)⁺: 695.3081, Found: 695.3096.

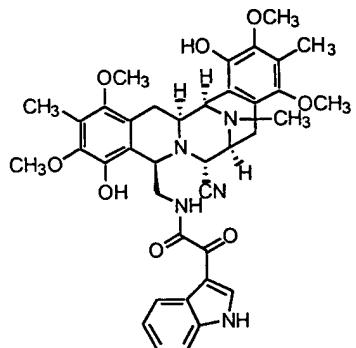
Example 15: 2-Furoic acid amide derivative



Diethylaniline (0.33 μ L, 2.1 μ mol, 1.1 equiv) was added in one portion to a stirred solution of the amine (1.0 mg, 1.9 μ mol, 1 equiv) in THF (0.15 mL) at 0 °C under an argon atmosphere and the solution was stirred for 10 min. 2-Furoic acid (0.28 mg, 2.5 μ mol, 1.3 equiv), 1-[3-(dimethylamino)propyl]-3-ethylcarbodiimide hydrochloride (0.48 mg, 2.5 μ mol, 1.3 equiv) and 1-hydroxybenzotriazole (0.31 mg, 2.3 μ mol, 1.2 equiv) were then added separately, each in one portion, to the above solution at 0 °C. The reaction mixture was warmed to 23 °C over 19 h, then was quenched with saturated aqueous ammonium chloride solution (3 mL). The mixture was diluted with ethyl acetate (10 mL) and the layers were separated. The aqueous layer was extracted with ethyl acetate (10 mL) and the combined organic layers were dried over sodium sulfate. Concentration in vacuo left a yellow oil, which was purified by flash column chromatography (70% ethyl acetate-hexanes) to give the 2-furoic acid amide derivative (1.0 mg, 85%) as a white solid.

$^1\text{H NMR}$ (500 MHz, CDCl_3), δ 7.18-7.17 (m, 1H, ArH), 6.70 (dd, 1H, J = 3.4, 1.0, ArH), 6.31 (dd, 1H, J = 3.4, 2.0, ArH), 6.07 (d, 1H, J = 6.9, NH), 5.63 (s, 1H, ArOH), 5.60 (s, 1H, ArOH), 4.28 (br. d, 1H, J = ~2.0, ArCHCH₂NH), 4.22 (d, 1H, J = 1.5, ArCHNCH₃), 4.10-4.05 (m, 2H, CHC≡N, ArCHCH₂NH), 3.83 (s, 3H, ArOCH₃), 3.74 (s, 3H, ArOCH₃), 3.54 (s, 3H, ArOCH₃), 3.45 (br. d, 1H, J = ~7.8, CHCHC≡N), 3.42 (s, 3H, ArOCH₃), 3.32 (ddd, 1H, J = 12.0, 2.7, 2.7, ArCHCHCH₂Ar), 3.23 (dd, 1H, J = 16.1, 2.4, ArCHCHCH₂Ar), 3.23-3.19 (1H, m, ArCHCH₂NH), 3.12 (dd, 1H, J = 18.6, 8.3, CH₂CHCHC≡N), 2.48 (d, 1H, J = 18.6, CH₂CHCHC≡N), 2.35 (s, 3H, NCH₃), 2.16 (s, 3H, ArCH₃), 2.15 (s, 3H, ArCH₃), 2.04 (dd, 1H, J = 16.1, 11.7, ArCHCHCH₂Ar). **FTIR** (neat film), cm^{-1} 3385, 2933, 2226, 1651, 1462. **HRMS (ES)** Calcd for $\text{C}_{33}\text{H}_{39}\text{N}_4\text{O}_8$ (MH^+): 619.2768, Found: 619.2790.

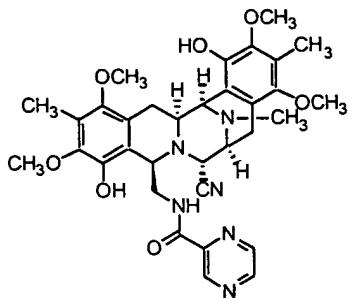
Example 16: Indole-3-glyoxamide derivative



5 Indole-3-glyoxylic acid (0.55 mg, 2.9 μ mol, 1.5 equiv) was added in one portion to a stirred solution of the amine (1.0 mg, 1.9 μ mol, 1 equiv) in dichloromethane (0.15 mL) at 23 °C under an argon atmosphere and the solution was stirred for 10 min. *N*-Cyclohexylcarbodiimide-*N'*-propyloxymethyl polystyrene (Argonaut Technologies, 1.13 mmol/g, 3.4 mg, 3.8 μ mol, 2.0 equiv) was then added, in one portion, to the above solution at 23 °C. The reaction mixture was
10 stirred gently at 23 °C for 15 h 20 min, then was purified by flash column chromatography (70% ethyl acetate-hexanes) to give the indole-3-glyoxamide derivative (0.9 mg, 60%) as a white solid.

1H NMR (500 MHz, CDCl₃), δ 8.85 (d, 1H, J = 3.4, ArH), 8.65 (br. s, 1H, ArNH), 8.20 (dd, 1H, J = 7.1, 2.1, ArH), 7.42 (dd, 1H, J = 6.6, 1.7, ArH), 7.32-7.28 (m, 2H, ArH), 7.01 (t, 1H, J = 5.7, NH), 5.67 (s, 1H, ArOH), 5.55 (s, 1H, ArOH), 4.31 (br. t, 1H, CHCH₂NHCO), 4.19
15 (d, 1H, J = 2.0, ArCHNCH₃), 4.10 (d, 1H, J = 2.4, CHC≡N), 3.76 (s, 3H, ArOCH₃), 3.75-3.71 (m, 4H, ArOCH₃, CH₂NHCO), 3.61 (s, 3H, ArOCH₃), 3.57 (s, 3H, ArOCH₃), 3.45-3.39 (m, 2H, CHCHC≡N, CH₂NHCO), 3.29-3.25 (m, 2H, ArCHCHCH₂Ar, ArCHCHCH₂Ar), 3.09 (dd, 1H, J = 18.4, 8.0, CH₂CHCHC≡N), 2.58 (d, 1H, J = 18.4, CH₂CHCHC≡N), 2.33 (s, 3H, NCH₃), 2.18 (s, 3H, ArCH₃), 2.10 (dd, 1H, J = 16.4, 12.0, ArCHCHCH₂Ar), 1.95 (s, 3H, ArCH₃). **FTIR** (neat film), cm⁻¹ 3370, 2926, 2227, 1675, 1625. **HRMS (ES)** Calcd for C₃₈H₄₁N₅O₈ (MH)⁺: 696.3033, Found: 696.3002.

Example 17: Pyrazine-2-carboxylic acid amide derivative

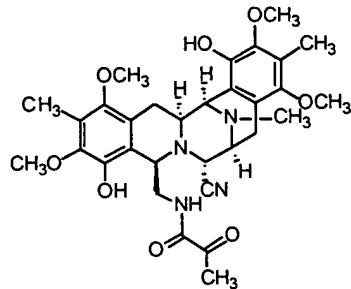


Diethylaniline (0.33 μ L, 2.1 μ mol, 1.1 equiv) was added in one portion to a stirred solution of the amine (1.0 mg, 1.9 μ mol, 1 equiv) in THF (0.2 mL) at 0 °C under an argon atmosphere and the solution was stirred for 5 min. 2-Pyrazinecarboxylic acid (0.31 mg, 2.5 μ mol, 1.3 equiv), 1-[3-(dimethylamino)propyl]-3-ethylcarbodiimide hydrochloride (0.48 mg, 2.5 μ mol, 1.3 equiv) and 1-hydroxybenzotriazole (0.31 mg, 2.3 μ mol, 1.2 equiv) were then added separately, each in one portion, to the above solution at 0 °C. The reaction mixture was warmed to 23 °C over 17 h 40 min, then was quenched with saturated aqueous ammonium chloride solution (3 mL). The mixture was diluted with ethyl acetate (10 mL) and the layers were separated. The aqueous layer was extracted with ethyl acetate (10 mL) and the combined organic layer was dried over sodium sulfate. Concentration in vacuo left a white solid, which was purified by flash column chromatography (80% ethyl acetate-hexanes) to give the pyrazine-2-carboxylic acid amide derivative (1.1 mg, 92%) as a white solid.

¹H NMR (500 MHz, CDCl₃), δ 9.10 (d, 1H, J = 1.5, ArH), 8.60 (d, 1H, J = 2.4, ArH), 8.17 (app. t, 1H, J = ~2.0, ArH), 7.33 (t, 1H, J = 5.4, NH), 5.65 (s, 1H, ArOH), 5.52 (s, 1H, ArOH), 4.33 (br. s, 1H, CHCH₂NHCO), 4.18 (br. s, 1H, ArCHNCH₃), 4.11 (d, 1H, J = 2.4, CHC≡N), 3.93 (ddd, 1H, J = 13.4, 6.7, 2.0, CH₂NHCO), 3.75 (s, 3H, ArOCH₃), 3.74 (s, 3H, ArOCH₃), 3.57 (s, 3H, ArOCH₃), 3.49 (ddd, 1H, J = 13.4, 4.4, 4.4, CH₂NHCO), 3.45 (obsc. d, 1H, CHCHC≡N), 3.44 (s, 3H, ArOCH₃), 3.29 (br. d, 1H, J = 11.7, ArCHCHCH₂Ar), 3.20 (dd, 1H, J = 16.1, 2.4, ArCHCHCH₂Ar), 3.07 (dd, 1H, J = 18.5, 8.3, CH₂CHCHC≡N), 2.55 (d, 1H, J = 18.5, CH₂CHCHC≡N), 2.32 (s, 3H, NCH₃), 2.16 (s, 3H, ArCH₃), 2.08 (dd, 1H, J = 16.1, 12.2, ArCHCHCH₂Ar), 2.08 (s, 3H, ArCH₃). FTIR (neat film), cm⁻¹ 3372, 2922, 2231, 1728, 1676. HRMS (ES) Calcd for C₃₃H₃₈N₆O₇ (MH)⁺: 631.2880, Found: 631.2853.

25

Example 18: Pyruvamide derivative

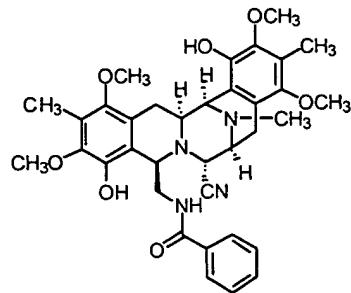


Diethylaniline (0.37 μ L, 2.3 μ mol, 1.1 equiv) was added in one portion to a stirred solution of the amine (1.1 mg, 2.1 μ mol, 1 equiv) in THF (0.15 mL) at 0 °C under an argon atmosphere and the solution was stirred for 5 min. Pyruvic acid (0.18 μ L, 2.7 μ mol, 1.3 equiv), 5 1-[3-(dimethylamino)propyl]-3-ethylcarbodiimide hydrochloride (0.52 mg, 2.7 μ mol, 1.3 equiv) and 1-hydroxybenzotriazole (0.34 mg, 2.5 μ mol, 1.2 equiv) were then added separately, each in one portion, to the above solution at 0 °C. The reaction mixture was warmed to 23 °C over 14 h 15 min, then was quenched with saturated aqueous ammonium chloride solution (3 mL). The 10 mixture was diluted with ethyl acetate (10 mL) and the layers were separated. The aqueous layer was extracted with ethyl acetate (10 mL) and the combined organic layer was dried over sodium sulfate. Concentration in vacuo left a white solid, which was purified by flash column chromatography (70% ethyl acetate-hexanes) to give the pyruvamide derivative (1.2 mg, 96%) as a white solid.

15 **^1H NMR** (500 MHz, CDCl_3), δ 6.45 (t, 1H, J = 5.6, NHCO), 5.62 (s, 1H, ArOH), 5.56 (s, 1H, ArOH), 4.24 (dd, 1H, J = 4.1, 1.9, CHCH₂NHCO), 4.17 (dd, 1H, J = 3.1, 1.2, ArCHNCH₃), 4.03 (d, 1H, J = 2.9, CHC≡N), 3.81 (s, 3H, ArOCH₃), 3.76 (s, 3H, ArOCH₃), 3.67 (s, 3H, ArOCH₃), 3.66 (app. ddd (obsc), 1H, CH₂NHCO), 3.60 (s, 3H, ArOCH₃), 3.42 (br. d, 1H, J = ~8.8, CHCHC≡N), 3.31 (ddd, 1H, J = 13.6, 5.6, 4.1, CH₂NHCO), 3.26-3.21 (m, 2H, 20 ArCHCHCH₂Ar, ArCHCHCH₂Ar), 3.07 (dd, 1H, J = 18.6, 8.3, CH₂CHCHC≡N), 2.49 (d, 1H, J = 18.6, CH₂CHCHC≡N), 2.32 (s, 3H, NCH₃), 2.24 (s, 3H, COCH₃), 2.20 (s, 3H, ArCH₃), 2.17 (s, 3H, ArCH₃), 1.93 (dd, 1H, J = 16.4, 12.0, ArCHCHCH₂Ar). **FTIR** (neat film), cm^{-1} 3378, 2937, 2251, 1720, 1682, 1463. **HRMS (ES)** Calcd for C₃₁H₃₉N₄O₈ (MH)⁺: 595.2768, Found: 595.2787.

25

Example 19: Benzamide derivative

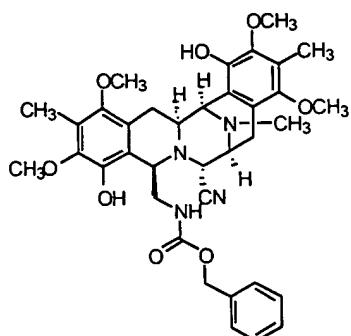


Diethylaniline (0.33 μ L, 2.1 μ mol, 1.1 equiv) and benzoyl chloride (0.66 μ L, 5.7 μ mol, 3.0 equiv) were added separately, each in one portion, to a stirred solution of the amine (1.0 mg,

5 1.9 μ mol, 1 equiv) in dichloromethane (0.15 mL) at 0 °C under an argon atmosphere. The reaction mixture was stirred at 0 °C for 30 min, then was quenched by the addition of a 1:1 mixture of water and saturated aqueous sodium hydrogen carbonate solution (4 mL). The mixture was extracted with ethyl acetate (2×15 mL) and the combined organic layer was washed with a 1:1 mixture of brine solution and saturated aqueous sodium hydrogen carbonate solution (4 mL). The organic layer was dried over sodium sulfate and concentrated in vacuo to leave a white solid. Purification by flash column chromatography (70% ethyl acetate-hexanes) gave the benzamide derivative (1.1 mg, 92%) as a white solid.

10 **^1H NMR** (500 MHz, CDCl_3), δ 7.39-7.36 (m, 1H, ArH), 7.23-7.20 (m, 2H, ArH), 7.06-
7.04 (m, 2H, ArH), 5.99 (br. d, 1H, $J = \sim 6.3$, NHCO), 5.67 (s, 1H, ArOH), 5.65 (s, 1H, ArOH),
15 4.31 (br. d, 1H, $J = \sim 2.4$, CHCH_2NHCO), 4.24 (dd, 1H, $J = 2.9, 1.5$, ArCHNCH₃), 4.20 (ddd,
1H, $J = 13.7, 8.3, 2.0$, CH_2NHCO), 4.10 (d, 1H, $J = 2.9$, $\text{CHC}\equiv\text{N}$), 3.82 (s, 3H, ArOCH₃), 3.74
(s, 3H, ArOCH₃), 3.45 (br. d, 1H, $J = \sim 8.8$, $\text{CHCHC}\equiv\text{N}$), 3.44 (s, 3H, ArOCH₃), 3.38 (s, 3H,
ArOCH₃), 3.36 (app. ddd (obsc), 1H, ArCHCHCH₂Ar), 3.32-3.27 (m, 2H, CH_2NHCO ,
ArCHCHCH₂Ar), 3.11 (dd, 1H, $J = 18.5, 7.8$, $\text{CH}_2\text{CHCHC}\equiv\text{N}$), 2.43 (d, 1H, $J = 18.5$,
20 $\text{CH}_2\text{CHCHC}\equiv\text{N}$), 2.33 (s, 3H, NCH₃), 2.16 (s, 3H, ArCH₃), 2.07 (s, 3H, ArCH₃), 2.03 (dd, 1H, $J = 15.9, 12.0$, ArCHCHCH₂Ar). **FTIR** (neat film), cm^{-1} 3395, 2933, 2226, 1656, 1462, 1056.
HRMS (ES) Calcd for $\text{C}_{35}\text{H}_{41}\text{N}_4\text{O}_7$ (MH^+): 629.2975, Found: 629.3002.

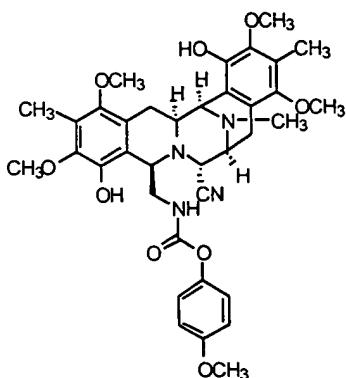
Example 20: Carbamic acid benzyl ester derivative



Diethylaniline (0.27 μ L, 1.7 μ mol, 1.1 equiv) and benzyl chloroformate (0.65 μ L, 4.6 μ mol, 3.0 equiv) were added separately, each in one portion, to a stirred solution of the amine (0.8 mg, 1.5 μ mol, 1 equiv) in dichloromethane (0.1 mL) at 0 °C under an argon atmosphere. The reaction mixture was stirred at 0 °C for 30 min, then was quenched by the addition of a 1:1 mixture of water and saturated aqueous sodium hydrogen carbonate solution (3 mL). The mixture was extracted with ethyl acetate (2×10 mL) and the combined organic layer was washed with a 1:1 mixture of brine solution and saturated aqueous sodium hydrogen carbonate solution (5 mL). The organic layer was dried over sodium sulfate and concentrated in vacuo to leave a light yellow oil. Purification by flash column chromatography (60% ethyl acetate-hexanes) gave the carbamic acid benzyl ester derivative (1.0 mg, 99%) as a white solid.

^1H NMR (500 MHz, CDCl_3), δ 7.30-7.27 (m, 3H, ArH), 7.14-7.12 (m, 2H, ArH), 5.58 (s, 1H, ArOH), 5.52 (s, 1H, ArOH), 4.80 (s, 2H, OCH_2Ar), 4.42 (br. d, 1H, $J = \sim 5.4$, NHCO), 4.17-4.15 (m, 2H, ArCHNCH₃, CHCH₂NHCO), 3.99 (d, 1H, $J = 2.0$, CHC≡N), 3.74 (s, 3H, ArOCH₃), 3.64 (s, 3H, ArOCH₃), 3.60 (s, 3H, ArOCH₃), 3.58 (app. ddd (obsc), 1H, CH₂NHCO), 3.58 (s, 3H, ArOCH₃), 3.38 (br. d, 1H, $J = \sim 7.3$, CHCHC≡N), 3.28 (ddd, 1H, $J = 12.0, 2.7, 2.7$, ArCHCHCH₂Ar), 3.20 (dd, 1H, $J = 15.8, 2.4$, ArCHCHCH₂Ar), 3.14 (ddd, 1H, $J = 13.4, 3.9, 3.9$, CH₂NHCO), 3.07 (dd, 1H, $J = 18.6, 7.8$, CH₂CHCHC≡N), 2.47 (d, 1H, $J = 18.6$, CH₂CHCHC≡N), 2.33 (s, 3H, NCH₃), 2.20 (s, 3H, ArCH₃), 2.10 (s, 3H, ArCH₃), 1.90 (dd, 1H, $J = 15.8, 12.0$, ArCHCHCH₂Ar). **FTIR** (neat film), cm^{-1} 3405, 2923, 2226, 1713, 1462. **HRMS (ES)** Calcd for C₃₆H₄₃N₄O₈ (MH)⁺: 659.3081, Found: 659.3110.

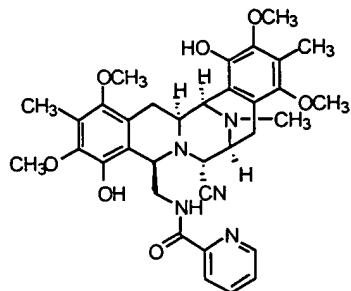
Example 21: Carbamic acid 4-methoxy-phenyl ester derivative



Diethylaniline (0.33 μ L, 2.1 μ mol, 1.1 equiv) and 4-methoxyphenyl chloroformate (0.85 μ L, 5.7 μ mol, 3.0 equiv) were added separately, each in one portion, to a stirred solution of the amine (1.0 mg, 1.9 μ mol, 1 equiv) in dichloromethane (0.15 mL) at 0 °C under an argon atmosphere. The reaction mixture was stirred at 0 °C for 40 min, then was quenched by the addition of a 1:1 mixture of water and saturated aqueous sodium hydrogen carbonate solution (3 mL). The mixture was extracted with ethyl acetate (2×10 mL) and the combined organic layer was washed with a 1:1 mixture of brine solution and saturated aqueous sodium hydrogen carbonate solution (5 mL). The organic layer was dried over sodium sulfate and concentrated in vacuo to leave a colorless oil. Purification by flash column chromatography (50% → 70% ethyl acetate-hexanes) gave the carbamic acid 4-methoxyphenyl ester derivative (0.9 mg, 70%) as a white solid.

$^1\text{H NMR}$ (500 MHz, CDCl_3), δ 6.75 (d, 1H, $J = 9.3$, ArH), 6.67 (d, 1H, $J = 8.8$, ArH), 5.61 (s, 1H, ArOH), 5.57 (s, 1H, ArOH), 4.67 (app. d, 1H, $J = 6.8$, NHCO), 4.21 (d, 1H, $J = 1.9$, ArCHNCH₃), 4.19 (br. d, 1H, $J = \sim 2.5$, CHCH₂NHCO), 4.05 (d, 1H, $J = 2.4$, CHC≡N), 3.75 (s, 3H, ArOCH₃), 3.74 (s, 3H, ArOCH₃), 3.71 (s, 3H, ArOCH₃), 3.67 (app. ddd (obsc), 1H, CH₂NHCO), 3.63 (s, 3H, ArOCH₃), 3.62 (s, 3H, ArOCH₃), 3.43 (br. d, 1H, $J = \sim 7.8$, CHCHC≡N), 3.34 (ddd, 1H, $J = 12.0, 2.5, 2.5$, ArCHCHCH₂Ar), 3.26 (dd, 1H, $J = 15.8, 2.8$, ArCHCHCH₂Ar), 3.16-3.10 (m, 2H, CH₂NHCO, CH₂CHCHC≡N), 2.51 (d, 1H, $J = 18.6$, CH₂CHCHC≡N), 2.37 (s, 3H, NCH₃), 2.22 (s, 3H, ArCH₃), 2.17 (s, 3H, ArCH₃), 2.01 (dd, 1H, $J = 15.8, 12.0$, ArCHCHCH₂Ar). FTIR (neat film), cm^{-1} 3508, 3385, 2923, 2226, 1733, 1195. HRMS (ES) Calcd for $\text{C}_{36}\text{H}_{43}\text{N}_4\text{O}_9$ (MH^+): 675.3030, Found: 675.3019.

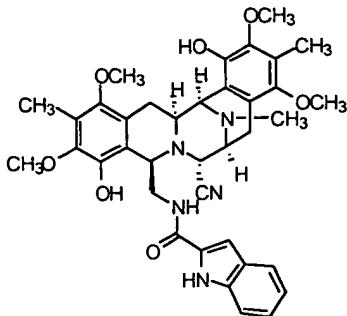
Example 22: Pyridine-2-carboxylic acid amide derivative



Diethylaniline (0.27 μ L, 1.7 μ mol, 1.1 equiv) was added in one portion to a stirred
5 solution of the amine (0.8 mg, 1.5 μ mol, 1 equiv) in THF (0.15 mL) at 0 °C under an argon
atmosphere and the solution was stirred for 10 min. Picolinic acid (0.24 mg, 2.0 μ mol, 1.3
equiv), 1-[3-(dimethylamino)propyl]-3-ethylcarbodiimide hydrochloride (0.38 mg, 2.0 μ mol, 1.3
equiv) and 1-hydroxybenzotriazole (0.25 mg, 1.8 μ mol, 1.2 equiv) were then added separately,
each in one portion, to the above solution at 0 °C. The reaction mixture was warmed to 23 °C
10 over 17 h 45 min, then was quenched with saturated aqueous ammonium chloride solution (3
mL). The mixture was diluted with ethyl acetate (10 mL) and the layers were separated. The
aqueous layer was extracted with ethyl acetate (10 mL) and the combined organic layer was
dried over sodium sulfate. Concentration in vacuo left a yellow oil, which was purified by flash
column chromatography (70% ethyl acetate-hexanes) to give the pyridine-2-carboxylic acid
15 amide derivative (1.0 mg, 100%) as a white solid.

$^1\text{H NMR}$ (500 MHz, CDCl_3), δ 8.20-8.19 (m, 1H, ArH), 7.92 (dd, 1H, J = 7.8, 1.0, ArH),
7.71 (ddd, 1H, J = 7.6, 7.6, 1.5, ArH), 7.68 (t, 1H, J = 5.6, NH), 7.30 (m, 1H, ArH), 5.65 (s, 1H,
ArOH), 5.52 (s, 1H, ArOH), 4.28 (t, 1H, J = 3.2, ArCH $_2$ NH), 4.22 (dd, 1H, J = 3.0, 1.0,
ArCHNCH $_3$), 4.13 (d, 1H, J = 2.5, CHC≡N), 3.85 (ddd, 1H, J = 13.2, 6.6, 3.0, ArCH $_2$ NH),
20 3.75 (s, 3H, ArOCH $_3$), 3.73 (s, 3H, ArOCH $_3$), 3.55 (s, 3H, ArOCH $_3$), 3.51-3.45 (m, 1H,
ArCH $_2$ NH), 3.44 (s, 3H, ArOCH $_3$), 3.44 (br. d, 1H, J = ~7.8, CHCHC≡N), 3.28 (ddd, 1H, J =
12.0, 3.0, 3.0, ArCHCHCH $_2$ Ar), 3.22 (dd, 1H, J = 15.7, 2.4, ArCHCHCH $_2$ Ar), 3.08 (dd, 1H, J =
18.6, 8.1, CH $_2$ CHCHC≡N), 2.58 (d, 1H, J = 18.6, CH $_2$ CHCHC≡N), 2.31 (s, 3H, NCH $_3$), 2.16 (s,
3H, ArCH $_3$), 2.16 (dd, 1H (obsc), ArCHCHCH $_2$ Ar), 2.09 (s, 3H, ArCH $_3$). **HRMS (ES)** Calcd
25 for $\text{C}_{34}\text{H}_{40}\text{N}_5\text{O}_7$ (MH^+): 630.2927, Found: 630.2904.

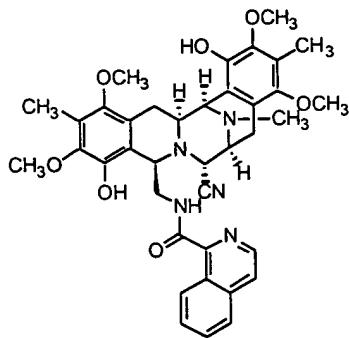
Example 23: Indole-2-carboxylic acid amide derivative



Diethylaniline (0.33 μ L, 2.1 μ mol, 1.1 equiv) was added in one portion to a stirred solution of the amine (1.0 mg, 1.9 μ mol, 1 equiv) in THF (0.15 mL) at 0 °C under an argon atmosphere and the solution was stirred for 5 min. Indole-2-carboxylic acid (0.40 mg, 2.5 μ mol, 1.3 equiv), 1-[3-(dimethylamino)propyl]-3-ethylcarbodiimide hydrochloride (0.48 mg, 2.5 μ mol, 1.3 equiv) and 1-hydroxybenzotriazole (0.31 mg, 2.3 μ mol, 1.2 equiv) were then added separately, each in one portion, to the above solution at 0 °C. The reaction mixture was warmed to 23 °C over 15 h 30 min, then was quenched with saturated aqueous ammonium chloride solution (3 mL). The mixture was diluted with ethyl acetate (10 mL) and the layers were separated. The aqueous layer was extracted with ethyl acetate (10 mL) and the combined organic layer was dried over sodium sulfate. Concentration in vacuo left a white solid, which was purified by flash column chromatography (60% ethyl acetate-hexanes) to give the indole-2-carboxylic acid amide derivative (1.1 mg, 87%) as a white solid.

$^1\text{H NMR}$ (500 MHz, CDCl_3), δ 8.80 (s, 1H, ArNH), 7.65 (d, 1H, J = 8.7, ArH), 7.31 (d, 1H, J = 8.3, ArH), 7.23 (ddd, 1H, J = 6.9, 6.9, 1.0, ArH), 7.12 (ddd, 1H, J = 6.9, 6.9, 1.0, ArH), 5.89 (dd, 1H, J = 8.3, 2.5, NH), 5.69 (s, 1H, ArOH), 5.68 (s, 1H, ArOH), 5.58 (d, 1H, J = 1.5, ArH), 4.31 (br. s, 1H, ArCH CH_2 NH), 4.26 (dd, 1H, J = 3.0, 1.0, ArCHNCH $_3$), 4.16 (ddd, 1H, J = 13.6, 8.5, 2.4, ArCH CH_2 NH), 4.10 (d, 1H, J = 2.5, CHC≡N), 3.80 (s, 3H, ArOCH $_3$), 3.74 (s, 3H, ArOCH $_3$), 3.56 (s, 3H, ArOCH $_3$), 3.48 (br. d, 1H, J = ~8.5, CHCHC≡N), 3.37-3.33 (m, 2H, ArCH CH_2 NH, ArCH CHCH_2 Ar), 3.32 (s, 3H, ArOCH $_3$), 3.25 (dd, 1H, J = 16.1, 2.9, ArCHCHCH $_2$ Ar), 3.14 (dd, 1H, J = 18.5, 8.3, CH $\text{CHCHC}\equiv\text{N}$), 2.55 (d, 1H, J = 18.5, CH $\text{CHCHC}\equiv\text{N}$), 2.35 (s, 3H, NCH $_3$), 2.15 (s, 3H, ArCH $_3$), 2.13 (s, 3H, ArCH $_3$), 2.01 (dd, 1H, J = 16.1, 12.2, ArCH CHCH_2 Ar). **FTIR** (neat film), cm^{-1} 3405, 3292, 2923, 2226, 1646, 1544, 1056. **HRMS (ES)** Calcd for C $_{37}\text{H}_{42}\text{N}_5\text{O}_7$ (MH^+): 668.3084, Found: 668.3112.

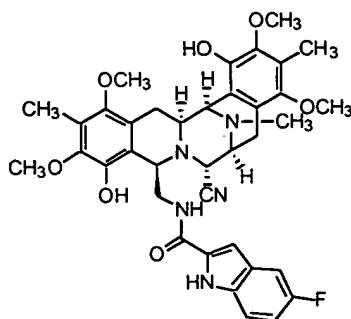
Example 24: Isoquinoline-1-carboxylic acid amide derivative



5 1-Isoquinolinecarboxylic acid (0.59 mg, 3.4 μ mol, 1.5 equiv) was added in one portion to
a stirred solution of the amine (1.2 mg, 2.3 μ mol, 1 equiv) in dichloromethane (0.15 mL) at 23
 $^{\circ}$ C under an argon atmosphere and the solution was stirred for 10 min. *N*-
10 Cyclohexylcarbodiimide-*N'*-propyloxymethyl polystyrene (Argonaut Technologies, 1.13
mmol/g, 4.0 mg, 4.6 μ mol, 2.0 equiv) was then added, in one portion, to the above solution at 23
 $^{\circ}$ C. The reaction mixture was stirred gently at 23 $^{\circ}$ C for 21 h, then was purified by flash column
15 chromatography (85% ethyl acetate-hexanes) to give the isoquinoline-1-carboxylic acid amide
derivative (1.1 mg, 71%) as a white solid.

10 1 H NMR (500 MHz, CDCl₃), δ 9.30 (d, 1H, *J* = 8.8, ArH), 8.09 (d, 1H, *J* = 5.4, ArH),
7.77 (d, 1H, *J* = 8.3, ArH), 7.76 (t, 1H, *J* = 6.4, NH), 7.68-7.65 (m, 2H, ArH), 7.60-7.56 (m, 1H,
15 ArH), 5.71 (s, 1H, ArOH), 5.55 (s, 1H, ArOH), 4.37 (t, 1H, *J* = 3.3, CHCH₂NHCO), 4.20 (app.
d, 1H, *J* = ~2.0, ArCHNCH₃), 4.16 (d, 1H, *J* = 2.5, CHC≡N), 3.92 (ddd, 1H, *J* = 13.6, 7.1, 3.2,
CH₂NHCO), 3.75 (s, 3H, ArOCH₃), 3.71 (s, 3H, ArOCH₃), 3.51 (ddd, 1H, *J* = 13.6, 4.4, 4.4,
CH₂NHCO), 3.44 (s, 4H, ArOCH₃, CHCHC≡N), 3.40 (s, 3H, ArOCH₃), 3.31 (ddd, 1H, *J* = 11.8,
2.7, 2.7, ArCHCHCH₂Ar), 3.24 (dd, 1H, *J* = 15.6, 2.4, ArCHCHCH₂Ar), 3.07 (dd, 1H, *J* = 18.4,
20 7.6, CH₂CHCHC≡N), 2.61 (d, 1H, *J* = 18.4, CH₂CHCHC≡N), 2.31 (s, 3H, NCH₃), 2.28 (dd, 1H,
J = 15.6, 11.7, ArCHCHCH₂Ar), 2.14 (s, 3H, ArCH₃), 1.89 (s, 3H, ArCH₃). FTIR (neat film),
cm⁻¹ 3376, 2926, 2230, 1665. HRMS (ES) Calcd for C₃₈H₄₁N₅O₇ (MH)⁺: 680.3084, Found:
680.3112.

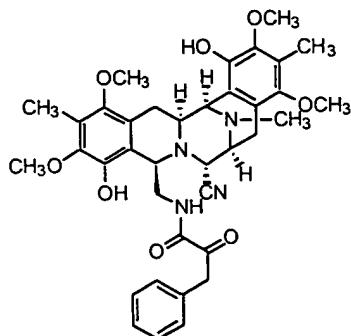
Example 25: 5-Fluoro-indole-2-carboxylic acid amide derivative



5-Fluoro-2-indolecarboxylic acid (0.51 mg, 2.9 μ mol, 1.5 equiv) was added in one
5 portion to a stirred solution of the amine (1.0 mg, 1.9 μ mol, 1 equiv) in dichloromethane (0.2 mL) at 23 °C under an argon atmosphere and the solution was stirred for 10 min. *N*-Cyclohexylcarbodiimide-*N'*-propyloxymethyl polystyrene (Argonaut Technologies, 1.13 mmol/g, 3.4 mg, 3.8 μ mol, 2.0 equiv) was then added, in one portion, to the above solution at 23 °C. The reaction mixture was stirred gently at 23 °C for 16 h 40 min, then was purified by flash 10 column chromatography (80% ethyl acetate-hexanes) to give the 5-fluoro-indole-2-carboxylic acid amide derivative (1.1 mg, 85%) as a white solid.

$^1\text{H NMR}$ (500 MHz, CDCl_3), δ 8.85 (br. s, 1H, ArNH), 7.32 (dd, 1H, J = 9.2, 2.4, ArH), 7.22 (dd, 1H, J = 10.8, 4.4, ArH), 6.98 (td, 1H, J = 8.8, 2.4, ArH), 5.87 (br. d, 1H, J = ~6.9, NH), 5.72 (s, 1H, ArOH), 5.69 (s, 1H, ArOH), 5.43 (br. d, 1H, J = ~2.0, ArH), 4.30 (br. s, 1H, CHCH_2NHCO), 4.25 (br. d, 1H, J = ~2.9, ArCHNCH₃), 4.20 (ddd, 1H, J = 13.4, 8.8, 2.0, CH_2NHCO), 4.09 (d, 1H, J = 2.5, $\text{CHC}\equiv\text{N}$), 3.84 (s, 3H, ArOCH₃), 3.73 (s, 3H, ArOCH₃), 3.55 (s, 3H, ArOCH₃), 3.47 (br. d, 1H, J = ~8.8, $\text{CHCHC}\equiv\text{N}$), 3.35 (ddd, 1H, J = 11.7, 2.7, 2.7, ArCHCHCH₂Ar), 3.28 (obsc. ddd, 1H, CH_2NHCO), 3.27 (s, 3H, ArOCH₃), 3.24 (dd, 1H, J = 16.1, 2.5, ArCHCHCH₂Ar), 3.13 (dd, 1H, J = 18.6, 8.3, $\text{CH}_2\text{CHCHC}\equiv\text{N}$), 2.53 (d, 1H, J = 18.6, $\text{CH}_2\text{CHCHC}\equiv\text{N}$), 2.35 (s, 3H, NCH₃), 2.15 (s, 3H, ArCH₃), 2.11 (s, 3H, ArCH₃), 1.98 (dd, 1H, J = 16.1, 12.0, ArCHCHCH₂Ar). **FTIR** (neat film), cm^{-1} 3398, 2925, 2225, 1645. **HRMS (ES)** Calcd for C₃₇H₄₀N₅O₇F (MH)⁺: 686.2990, Found: 686.3020.

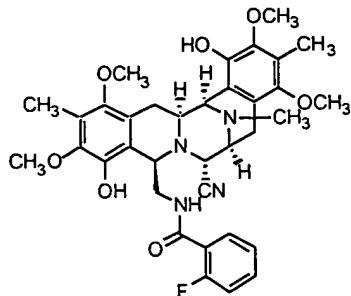
Example 26: Phenylpyruvamide derivative



Diethylaniline (0.43 μ L, 2.7 μ mol, 1.1 equiv) was added in one portion to a stirred solution of the amine (1.3 mg, 2.5 μ mol, 1 equiv) in THF (0.25 mL) at 0 °C under an argon atmosphere and the solution was stirred for 5 min. Phenylpyruvic acid (0.53 mg, 3.2 μ mol, 1.3 equiv), 1-[3-(dimethylamino)propyl]-3-ethylcarbodiimide hydrochloride (0.62 mg, 3.2 μ mol, 1.3 equiv) and 1-hydroxybenzotriazole (0.40 mg, 3.0 μ mol, 1.2 equiv) were then added separately, each in one portion, to the above solution at 0 °C. The reaction mixture was warmed to 23 °C over 13 h 40 min, then was quenched with saturated aqueous ammonium chloride solution (3 mL). The mixture was diluted with ethyl acetate (10 mL) and the layers were separated. The aqueous layer was extracted with ethyl acetate (10 mL) and the combined organic layer was dried over sodium sulfate. Concentration in vacuo left a white solid, which was purified by flash column chromatography (70% ethyl acetate-hexanes) to give the phenylpyruvamide derivative (1.4 mg, 84%) as a white solid.

^1H NMR (500 MHz, CDCl_3), δ 7.27-7.23 (m, 3H, ArH), 7.04-7.02 (m, 2H, ArH), 6.56 (br. d, 1H, $J = \sim 4.9$, NH), 5.61 (s, 1H, ArOH), 5.54 (s, 1H, ArOH), 4.25 (br. d, 1H, $J = \sim 2.0$, CHCH_2NHCO), 4.17 (br. d, 1H, $J = \sim 2.4$, ArCHNCH₃), 4.00 (d, 1H, $J = 2.4$, $\text{CHC}\equiv\text{N}$), 3.90 (AB system, 2H, ArCH₂COCONH), 3.84 (obsc. ddd, 1H, $J = 13.7, 8.5, 1.9$, CH_2NHCO), 3.74 (s, 3H, ArOCH₃), 3.60 (s, 3H, ArOCH₃), 3.54 (s, 3H, ArOCH₃), 3.52 (s, 3H, ArOCH₃), 3.41 (br. d, 1H, $J = \sim 7.8$, CHCHC≡N), 3.27 (ddd, 1H, $J = 11.7, 2.5, 2.5$, ArCHCHCH₂Ar), 3.21 (dd, 1H, $J = 16.1, 2.4$, ArCHCHCH₂Ar), 3.17 (ddd, 1H, $J = 13.7, 3.9, 3.9$, CH_2NHCO), 3.07 (dd, 1H, $J = 18.6, 8.3$, $\text{CH}_2\text{CHCHC}\equiv\text{N}$), 2.42 (d, 1H, $J = 18.6$, $\text{CH}_2\text{CHCHC}\equiv\text{N}$), 2.32 (s, 3H, NCH₃), 2.18 (s, 3H, ArCH₃), 2.11 (s, 3H, ArCH₃), 1.94 (dd, 1H, $J = 16.1, 12.0$, ArCHCHCH₂Ar). **FTIR** (neat film), cm^{-1} 3367, 2929, 2229, 1723, 1681. **HRMS (ES)** Calcd for $\text{C}_{37}\text{H}_{42}\text{N}_4\text{O}_8$ (MH^+): 671.3081, Found: 671.3112.

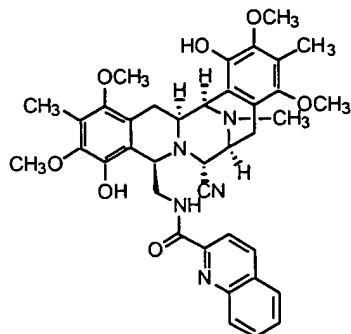
Example 27: 2-Fluorobenzamide derivative



2-Fluorobenzoic acid (0.36 mg, 2.6 μ mol, 1.5 equiv) was added in one portion to a stirred
5 solution of the amine (0.9 mg, 1.7 μ mol, 1 equiv) in dichloromethane (0.2 mL) at 23 °C under an argon atmosphere and the solution was stirred for 10 min. *N*-Cyclohexylcarbodiimide-*N'*-propyloxymethyl polystyrene (Argonaut Technologies, 1.13 mmol/g, 3.0 mg, 3.4 μ mol, 2.0 equiv) was then added, in one portion, to the above solution at 23 °C. The reaction mixture was stirred gently at 23 °C for 13 h, then was purified by flash column chromatography (70% ethyl
10 acetate-hexanes) to give the 2-fluorobenzamide derivative (1.1 mg, 99%) as a white solid.

^1H NMR (500 MHz, CDCl_3), δ 7.78 (td, 1H, J = 7.8, 1.9, ArH), 7.38-7.34 (m, 1H, ArH), 7.13 (td, 1H, J = 7.8, 1.0, ArH), 6.85-6.81 (m, 1H, ArH), 6.35 (br. s, 1H, NH), 5.68 (s, 1H, ArOH), 5.56 (s, 1H, ArOH), 4.32 (br. d, 1H, J = ~1.9, CHCH_2NHCO), 4.20 (dd, 1H, J = 12.9, 1.0, ArCHNCH₃), 4.12 (d, 1H, J = 2.5, CHC≡N), 4.09-4.05 (m, 1H, CH_2NHCO), 3.75 (s, 6H, 2
15 × ArOCH₃), 3.59 (s, 3H, ArOCH₃), 3.46 (obsc. ddd, 1H, J = ~14.2, 3.7, 3.7, CH_2NHCO), 3.44 (br. d, 1H, J = ~8.8, CHCHC≡N), 3.37 (s, 3H, ArOCH₃), 3.30 (br. d, 1H, J = ~11.7, ArCHCHCH₂Ar), 3.26 (dd, 1H, J = 15.6, 2.7, ArCHCHCH₂Ar), 3.06 (dd, 1H, J = 18.5, 8.0, CH₂CHCHC≡N), 2.43 (d, 1H, J = 18.5, $\text{CH}_2\text{CHCHC}\equiv\text{N}$), 2.30 (s, 3H, NCH₃), 2.19 (s, 3H, ArCH₃), 2.11 (dd, 1H, J = 15.6, 11.7, ArCHCHCH₂Ar), 2.00 (s, 3H, ArCH₃). **FTIR** (neat film),
20 cm⁻¹ 3403, 2932, 2231, 1649. **HRMS (ES)** Calcd for C₃₅H₃₉N₄O₇F (MH)⁺: 647.2881, Found: 647.2903.

Example 28: Quinoline-2-carboxylic acid amide derivative

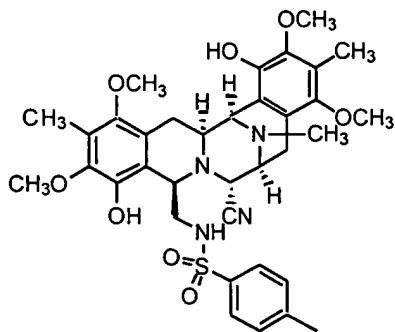


Quinaldic acid (0.50 mg, 2.9 μ mol, 1.5 equiv) was added in one portion to a stirred solution of the amine (1.0 mg, 1.9 μ mol, 1 equiv) in dichloromethane (0.2 mL) at 23 °C under an argon atmosphere and the solution was stirred for 10 min. *N*-Cyclohexylcarbodiimide-*N'*-propyloxymethyl polystyrene (Argonaut Technologies, 1.13 mmol/g, 3.4 mg, 3.8 μ mol, 2.0 equiv) was then added, in one portion, to the above solution at 23 °C. The reaction mixture was stirred gently at 23 °C for 16 h 50 min, then was purified by flash column chromatography (70% ethyl acetate-hexanes) to give the quinoline-2-carboxylic acid amide derivative (1.1 mg, 85%) as a white solid.

¹H NMR (500 MHz, CDCl₃), δ 8.23 (d, 1H, *J* = 8.3, ArH), 8.13 (d, 1H, *J* = 8.3, ArH), 7.95 (t, 1H, *J* = 5.6, NH), 7.84 (d, 1H, *J* = 8.1, ArH), 7.81 (d, 1H, *J* = 8.1, ArH), 7.72 (ddd, 1H, *J* = 8.3, 6.8, 1.4, ArH), 7.59 (ddd, 1H, *J* = 8.3, 6.8, 1.0, ArH), 5.71 (br. s, 1H, ArOH), 5.44 (s, 1H, ArOH), 4.39 (t, 1H, *J* = 3.9, CHCH₂NHCO), 4.19 (d, 1H, *J* = 2.4, CHC≡N), 4.17 (d, 1H, *J* = 1.5, ArCHNCH₃), 3.81-3.77 (m, 4H, CH₂NHCO, ArOCH₃), 3.61-3.57 (s, 7H, CH₂NHCO, 2 × ArOCH₃), 3.42 (br. d, 1H, CHCHC≡N), 3.38 (s, 3H, ArOCH₃), 3.31-3.26 (m, 2H, ArCHCHCH₂Ar, CH₂CHCHC≡N), 3.08 (dd, 1H, *J* = 18.6, 8.3, CH₂CHCHC≡N), 2.74 (d, 1H, *J* = 18.6, CH₂CHCHC≡N), 2.27 (s, 3H, NCH₃), 2.21 (s, 3H, ArCH₃), 2.16 (dd, 1H, *J* = 16.1, 12.2, ArCHCHCH₂Ar), 1.94 (s, 3H, ArCH₃). **FTIR** (neat film), cm⁻¹ 3385, 2933, 2226, 1667.

HRMS (ES) Calcd for C₃₈H₄₁N₅O₇ (MH)⁺: 680.3084, Found: 680.3094.

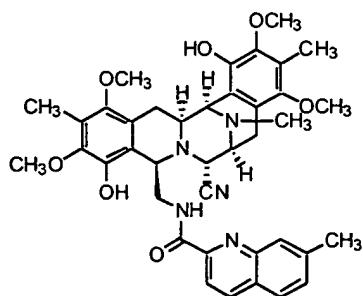
Example 29: 4-Methylphenyl sulfonamide derivative



5 Diethylaniline (0.33 μ L, 2.1 μ mol, 1.1 equiv) was added in one portion to a stirred solution of the amine (1.0 mg, 1.9 μ mol, 1 equiv) in THF (0.1 mL) at 0 °C under an argon atmosphere and the solution was stirred for 5 min. *p*-Toluenesulfonyl chloride (0.73 mg, 3.8 μ mol, 2.0 equiv) was then added in one portion to the above solution at 0 °C. The reaction mixture was stirred at 0 °C for 1 hr and then quenched with a 1:1 mixture of saturated aqueous 10 sodium hydrogen carbonate solution and water (4 mL). The mixture was diluted with ethyl acetate (10 mL) and the layers were separated. The aqueous layer was extracted with ethyl acetate (10 mL) and the combined organic layer was dried over sodium sulfate. Concentration in vacuo left a yellow oil, which was purified by flash column chromatography (60% ethyl acetate-hexanes) to give the 4-methylphenyl sulfonamide derivative (1.2 mg, 93%) as a white solid.

15

Example 30: 7-Methylquinoline-2-carboxylic acid amide derivative:



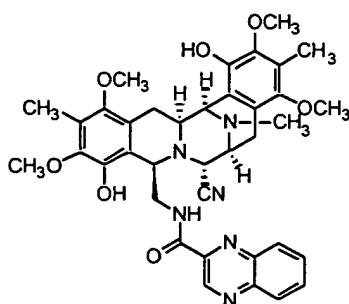
20 A solution of the amine 4 (1.0 mg, 1.9 μ mol, 1.0 equiv) in dichloromethane (0.1 mL) was added to 7-methylquinoline-2-carboxylic acid (2.8 μ mol, 1.5 equiv) and 1-hydroxybenzotriazole

(0.44 mg, 3.2 μ mol, 1.7 equiv) in a vial at 23 °C. The solution was stirred for 5 min, then *N*-cyclohexylcarbodiimide-*N'*-propyloxymethyl polystyrene (Argonaut Technologies, 1.13 mmol/g, 3.4 mg, 3.8 μ mol, 2.0 equiv) was added in one portion at 23 °C. The reaction mixture was stirred gently at 23 °C under an argon atmosphere for 16 h. PS-Trisamine (Argonaut Technologies, 4.71 mmol/g, 2.0 mg, 9.5 μ mol, 5.0 equiv) was then added in one portion and the reaction mixture was stirred for a further 2 h. The mixture was filtered through Celite (0.5 cm) and concentrated in vacuo to give the 7-methylquinoline-2-carboxylic acid amide derivative (1.2 mg, 95%) as an off-white solid.

¹H NMR (500 MHz, CDCl₃), δ 8.18 (d, 1H, *J* = 8.8, ArH), 8.07 (d, 1H, *J* = 8.3, ArH), 7.93 (br t, 1H, *J* = 5.8, NH), 7.73 (d, 1H, *J* = 8.3, ArH), 7.62 (s, 1H, ArH), 7.43 (dd, 1H, *J* = 8.3, 1.5, ArH), 5.72 (br s, 1H, ArOH), 5.42 (br s, 1H, ArOH), 4.39 (t, 1H, *J* = 3.9, CHCH₂NHCO), 4.19 (d, 1H, *J* = 2.9, CHC≡N), 4.16 (d, 1H, *J* = 1.5, ArCHNCH₃), 3.81–3.77 (m, 4H, CH₂NHCO, ArOCH₃), 3.64 (s, 3H, ArOCH₃), 3.58 (s, 3H, ArOCH₃), 3.51 (ddd, 1H, *J* = 13.2, 4.4, 4.4, CH₂NHCO), 3.42 (br d, 1H, *J* = 8.8, CHCHC≡N), 3.36 (s, 3H, ArOCH₃), 3.32–3.25 (m, 2H, ArCHCHCH₂Ar, ArCHCHCH₂Ar), 3.07 (dd, 1H, *J* = 18.1, 8.3, CH₂CHCHC≡N), 2.75 (d, 1H, *J* = 18.1, CH₂CHCHC≡N), 2.59 (s, 3H, ArCH₃), 2.28 (s, 3H, NCH₃), 2.23 (s, 3H, ArCH₃), 2.14 (dd, 1H, *J* = 15.4, 11.5, ArCHCHCH₂Ar), 1.92 (s, 3H, ArCH₃); FTIR (neat film, cm⁻¹) 3383 (m, br, OH/NH), 2934 (m), 2228 (w, C≡N), 1668 (s, C=O), 1463 (s); HRMS (ES) Calcd for C₃₉H₄₃N₅O₇ (MH)⁺: 694.3240, found: 694.3219.

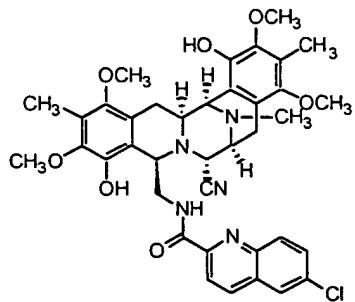
20

Example 31: Quinoxaline-2-carboxylic acid amide derivative:



¹H NMR (500 MHz, CDCl₃), δ 9.47 (s, 1H, ArH), 8.14 (d, 1H, J = 8.3, ArH), 7.85–7.82 (m, 1H, ArH), 7.79–7.76 (m, 2H, ArH), 7.53 (dd, 1H, J = 6.6, 4.6, NH), 5.69 (s, 1H, ArOH), 5.35 (s, 1H, ArOH), 4.39 (t, 1H, J = 3.2, CHCH₂NHCO), 4.18 (d, 1H, J = 2.4, CHC≡N), 4.13 (d, 1H, J = 1.9, ArCHNCH₃), 3.88 (ddd, 1H, J = 13.7, 7.6, 4.6, CH₂NHCO), 3.79 (s, 3H, ArOCH₃),
5 3.64 (1H, ddd, J = 13.7, 4.3, 3.0, CH₂NHCO), 3.61 (3H, s, ArOCH₃), 3.60 (3H, s, ArOCH₃), 3.44 (br d, 1H, J = 8.6, CHCHC≡N), 3.29–3.25 (m, 2H, ArCHCH₂Ar, ArCHCHCH₂Ar), 3.25 (s, 3H, ArOCH₃), 3.04 (dd, 1H, J = 18.5, 8.3, CH₂CHCHC≡N), 2.70 (d, 1H, J = 18.5, CH₂CHCHC≡N), 2.25 (s, 3H, NCH₃), 2.23 (s, 3H, ArCH₃), 2.12 (dd, 1H, J = 16.1, 12.2, ArCHCHCH₂Ar), 1.88 (s, 3H, ArCH₃); FTIR (neat film, cm⁻¹) 3383 (m, br, OH/NH), 2933 (m),
10 2228 (w, C≡N), 1673 (s, C=O), 1463 (s); HRMS (ES) Calcd for C₃₇H₄₀N₆O₇ (MH)⁺: 681.3036, found: 681.3062.

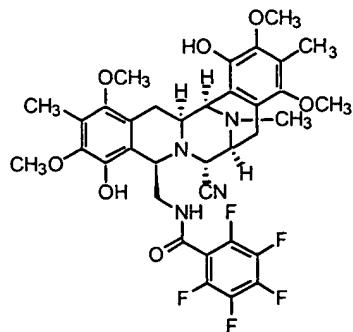
Example 32: 6-Chloroquinoline-2-carboxylic acid amide derivative:



15

¹H NMR (500 MHz, CDCl₃), δ 8.14 (s, 2H, ArH), 7.85 (t, 1H, J = 5.6, NH), 7.82 (d, 1H, J = 2.0, ArH), 7.74 (d, 1H, J = 9.2, ArH), 7.67 (dd, 1H, J = 9.3, 2.4, ArH), 5.69 (s, 1H, ArOH), 5.45 (s, 1H, ArOH), 4.39 (t, 1H, J = 3.7, CHCH₂NHCO), 4.17 (br s, 2H, CHC≡N, ArCHNCH₃), 3.76 (s, 3H, ArOCH₃), 3.72 (ddd, 1H, J = 13.7, 6.3, 4.4, CH₂NHCO), 3.64 (ddd, 1H, J = 13.7, 5.3, 3.9, CH₂NHCO), 3.56 (s, 3H, ArOCH₃), 3.54 (s, 3H, ArOCH₃), 3.49 (s, 3H, ArOCH₃), 3.43 (br d, 1H, J = 8.3, CHCHC≡N), 3.30–3.27 (m, 2H, ArCHCH₂Ar, ArCHCHCH₂Ar), 3.07 (dd, 1H, J = 18.3, 8.3, CH₂CHCHC≡N), 2.69 (d, 1H, J = 18.3, CH₂CHCHC≡N), 2.28 (s, 3H, NCH₃), 2.20 (s, 3H, ArCH₃), 2.16 (obs dd, 1H, J = 16.2, 11.7, ArCHCHCH₂Ar), 1.95 (s, 3H, ArCH₃); FTIR (neat film, cm⁻¹) 3383 (m, br, OH/NH), 2923 (s), 2228 (w, C≡N), 1668 (s, C=O), 1458 (s);
25 HRMS (ES) Calcd for C₃₈H₄₀N₅O₇Cl (MH)⁺: 714.2694, found: 714.2721.

Example 33: Pentafluorobenzamide derivative:

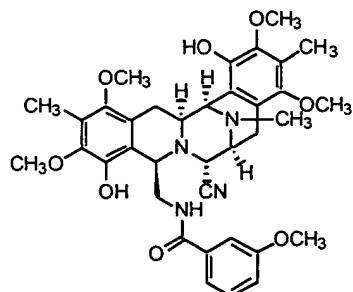


5

¹H NMR (500 MHz, CDCl₃), δ 5.66–5.65 (br m, 2H, ArOH, NH), 5.57 (s, 1H, ArOH), 4.27 (br m, 1H, CHCH₂NHCO), 4.17 (d, 1H, J = 1.5, ArCHNCH₃), 4.05–4.01 (m, 2H, CHC≡N, CH₂NHCO), 3.76 (s, 3H, ArOCH₃), 3.74 (s, 3H, ArOCH₃), 3.64 (s, 3H, ArOCH₃), 3.57 (s, 3H, ArOCH₃), 3.44 (br d, 1H, J = 8.3, CHCHC≡N), 3.34–3.30 (m, 2H, ArCHCHCH₂Ar, CH₂NHCO), 3.25 (dd, 1H, J = 16.2, 2.9, ArCHCHCH₂Ar), 3.05 (dd, 1H, J = 18.5, 7.8, CH₂CHCHC≡N), 2.43 (d, 1H, J = 18.5, CH₂CHCHC≡N), 2.32 (s, 3H, NCH₃), 2.21 (s, 3H, ArCH₃), 2.04 (s, 3H, ArCH₃), 1.95 (dd, 1H, J = 16.2, 12.2, ArCHCHCH₂Ar); FTIR (neat film, cm⁻¹) 3363 (m, br, OH/NH), 2933 (m), 2228 (w, C≡N), 1678 (s, C=O), 1502 (s); HRMS (ES) Calcd for C₃₅H₃₅N₄O₇F₅ (MH)⁺: 719.2504, found: 719.2479.

15

Example 34: 3-Methoxybenzamide derivative:



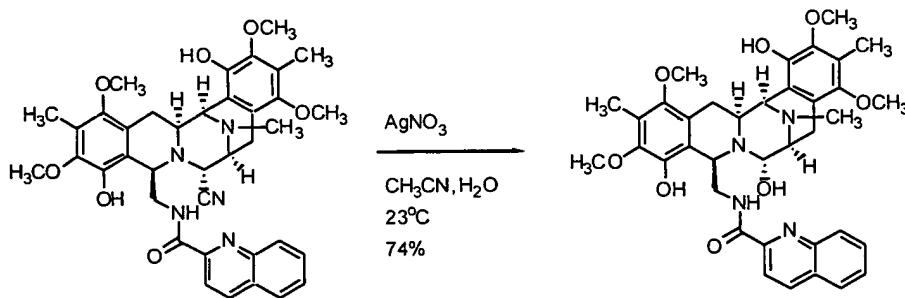
20

¹H NMR (500 MHz, CDCl₃), δ 7.10 (br s, 1H, ArH), 7.07 (t, 1H, J = 7.8, ArH), 6.91 (dd, 1H, J = 8.3, 1.9, ArH), 6.19 (d, 1H, J = 7.8, ArH), 6.00 (d, 1H, J = 5.9, NH), 5.69 (br s, 1H,

ArOH), 5.64 (s, 1H, ArOH), 4.31 (br d, 1H, J = 2.4, CHCH₂NHCO), 4.23 (d, 1H, J = 2.0, ArCHNCH₃), 4.13 (obs ddd, 1H, J = 13.7, 8.3, 2.0, CH₂NHCO), 4.10 (d, 1H, J = 2.4, CHC≡N), 3.79 (s, 3H, ArOCH₃), 3.75 (s, 3H, ArOCH₃), 3.74 (s, 3H, ArOCH₃), 3.48 (s, 3H, ArOCH₃), 3.45 (br d, 1H, J = 8.8, CHCHC≡N), 3.42 (s, 3H, ArOCH₃), 3.36–3.27 (m, 3H, ArCHCH₂Ar, 5 CH₂NHCO (1H), ArCHCHCH₂Ar (1H)), 3.10 (dd, 1H, J = 18.6, 8.3, CH₂CHCHC≡N), 2.43 (d, 1H, J = 18.6, CH₂CHCHC≡N), 2.32 (s, 3H, NCH₃), 2.16 (s, 3H, ArCH₃), 2.08 (s, 3H, ArCH₃), 2.03 (dd, 1H, J = 16.1, 11.7, ArCHCHCH₂Ar); FTIR (neat film, cm⁻¹) 3395 (m, br, OH/NH), 2933 (m), 2226 (w, C≡N), 1651 (m, C=O), 1462 (s); HRMS (ES) Calcd for C₃₆H₄₂N₄O₈ (MH)⁺: 659.3081, found: 659.3081.

10

Example 35: Hemiaminal derivative

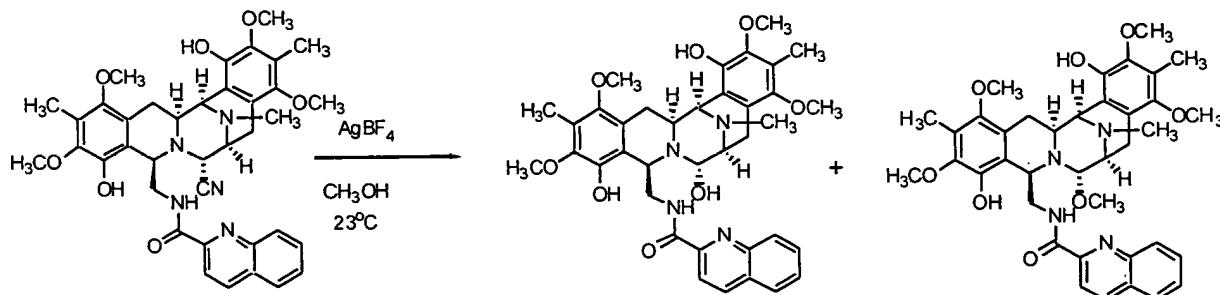


A solution of silver nitrate (15.9 mg, 93.6 μmol, 58.5 equiv) in a mixture of water and 15 acetonitrile (3:2, 500 μl) was added to the quinoline-2-carboxylic acid amide derivative (1.1 mg, 1.6 μmol, 1 equiv). The reaction mixture was stirred at 23 °C in the dark for 4 h. A 1:1 mixture of saturated aqueous sodium bicarbonate and saturated aqueous sodium chloride solution (2 mL, freshly prepared) was added and the mixture was stirred for 5 min. A further 1:1 mixture of saturated aqueous sodium bicarbonate and saturated aqueous sodium chloride solution (20 mL, freshly prepared) was then added. The mixture was extracted with methylene chloride (5x10 ml). The combined organic extract was dried over sodium sulfate and was then concentrated to leave a solid residue. The solid residue was purified by flash column chromatography (4% methanol-methylene chloride) to provide the hemiaminal (0.8 mg, 74%) as a white solid.

25 ¹H NMR (500 MHz, CDCl₃), δ 8.22 (d, 1H, J = 8.5, ArH), 8.12 (d, 1H, J = 8.5, ArH), 8.04 (t, 1H, J = 5.5, NH), 7.82 (d, 1H, J = 8.5, ArH), 7.72 (m, 2H, ArH), 7.59 (ddd, 1H, J = 8.5,

4.5, 1.5, ArH), 5.72 (br s, 1H, ArOH), 5.54 (s, 1H, ArOH), 4.84 (t, 1H, J = 4.0, CHCH₂NHCO), 4.60 (d, 1H, J = 2.0, CHOH), 4.12 (dd, 1H, J = 3.5, 1.5, CHCHOH), 3.78-3.68 (m, 4H, CH₂NHCO, ArOCH₃), 3.63 (1s, 3H, ArOCH₃), 3.47 (1s, 3H, ArOCH₃), 3.45 (1s, 3H, ArOCH₃). 3.38 (m, 1H, CH₂NHCO), 3.27 (m, 2H, ArCHCHCH₂Ar, ArCHCHCH₂Ar), 3.01 (dd, 1H, J = 5 16.5, 8.0, ArCHCHCH₂Ar), 2.60 (d, 2H, J = 17, CH₂CHCHOH), 2.27 (s, 3H, NCH₃), 2.21 (dd, 1H, J = 16.5, 11.5, ArCHCHCH₂Ar), 2.17 (1s, 3H, ArCH₃), 1.92 (1s, 3H, ArCH₃). FTIR (neat film), cm⁻¹ 3360, 2936, 1667, 1415. HRMS (ES) Calcd for C₃₇H₄₃N₄O₈ (MH)⁺: 671.3081, Found: 671.3083.

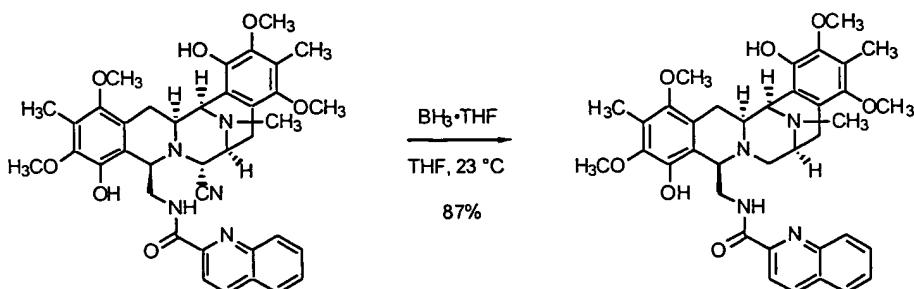
10 Example 36: Methoxy derivative



A solution of the quinoline-2-carboxylic acid amide derivative (0.8 mg, 1.2 μ mol, 1 equiv) in methanol (0.5 mL) was added, via cannula, to silver tetrafluoroborate (22 mg, 113 μ mol, 96 equiv) in an oven-dried glass vial. The reaction mixture was stirred at 23 °C in the dark for 24 h. A 1:1 mixture of saturated aqueous sodium bicarbonate and saturated aqueous sodium chloride solution (1 mL, freshly prepared) was added and the mixture was stirred for 5 min. A further 1:1 mixture of saturated aqueous sodium bicarbonate and saturated aqueous sodium chloride solution (10 mL, freshly prepared) was then added. The mixture was extracted with methylene chloride (4x10 mL). The combined organic extract was dried over sodium sulfate and was concentrated. The solid residue was purified by flash column chromatography (4% methanol-methylene chloride) to provide the hemiaminal (0.45 mg, 57%) as a white solid and the methoxy derivative (0.16 mg, 19.8%) as a white solid.

¹H NMR of the methoxy derivative (500 MHz, CDCl₃), δ 8.19 (d, 1H, J = 8.0, ArH), 8.07 (d, 1H, J = 8.0, ArH), 7.83 (m, 2H, NH, ArH), 7.76 (t, 1H, J = 7.0, ArH), 7.62 (t, 1H, J = 7.0, ArH), 7.53 (d, 1H, J = 8.0, ArH), 5.60 (br s, 1H, ArOH), 5.33 (s, 1H, ArOH), 4.64 (s, 1H, CHCH₂NHCO), 4.59 (d, 1H, J = 3.0, CHOCH₃), 4.18 (d, 1H, J = 3.5, 1.5, CH₂NHCO), 4.06 (dd, 5 1H, J = 14, 7.5, CHCHOH), 3.78 (1s, 3H, ArOCH₃), 3.69 (1s, 3H, ArOCH₃), 3.59 (m, 1H, CH₂NHCO), 3.37 (m, 1H, ArCHCHCH₂Ar), 3.29 (m, 1H, ArCHCHCH₂Ar), 3.16 (1s, 3H, ArOCH₃), 3.05 (1s, 3H, ArOCH₃), 2.99 (dd, 1H, J = 18.5, 9, CH₂CHCHOCH₃), 2.88 (d, 1H, J = 9, CH₂CHCHOCH₃), 2.52 (s, 3H, CHOCH₃), 2.36 (d, J = 18.5, ArCHCHCH₂Ar), 2.27 (s, 3H, NCH₃), 1.96 (ddd, J = 18.5, 11, 2.5, ArCHCHCH₂Ar), 1.76 (1s, 3H, ArCH₃), 1.74 (1s, 3H, 10 ArCH₃). HRMS (ES) Calcd for C₃₈H₄₅N₄O₈ (MH)⁺: 685.3237, Found: 685.3259.

Example 37: Reduced quinalascidin analog:



15

A solution of borane tetrahydrofuran complex in tetrahydrofuran (1.0 M, 9.0 μL, 9 μmol, 5.0 equiv) was added to a solution of the quinoline-2-carboxylic acid amide derivative (1.2 mg, 1.8 μmol, 1 equiv) in tetrahydrofuran (0.2 mL). The resulting colorless solution was stirred at 23 °C for 2.5 h. 1,4-Diazabicyclo[2.2.2]octane (DABCO, 6.1 mg, 0.054 mmol, 30 equiv) and water (0.2 mL) were added to the reaction mixture in sequence. The resulting colorless solution was stirred at 23 °C for 20 h. The solution was diluted with dichloromethane (5 mL) and the resulting mixture was dried over sodium sulfate and was concentrated. Purification of the white solid residue by flash chromatography (10% methanol–distilled ethyl acetate) afforded the reduced derivative (1.0 mg, 87%) as a white solid: *R*_f 0.16, 10% methanol–ethyl acetate; ¹H NMR (500 MHz, CD₃OD) δ 8.17 (d, 1H, J = 8.5 Hz, CHCHCCONH), 8.08 (d, 1H, J = 8.5 Hz, CHCHCCONH), 8.05 (br t, 1H, CONH), 7.80 (d, 1H, J = 8.5 Hz, NCCH), 7.73 (ddd, 1H, J = 7.5, 6.8, 1.5 Hz, NCCHCH), 7.70 (d, 1H, J = 8.0 Hz, NCCHCHCHCH), 7.57 (ddd, 1H, J = 8.0, 25 7.5, 6.8 Hz, NCCHCHCH).

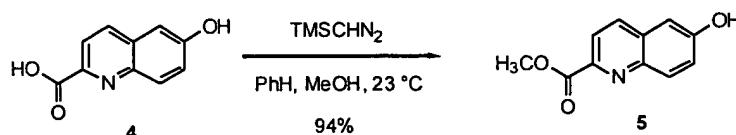
7.0, 1.5 Hz, NCCHCHCH), 5.75 (br s, 1H, ArOH), 5.55 (s, 1H, ArOH), 4.20 (br s, 1H, ArCHNCH₃), 4.02 (br t, 1H, CHCH₂NHCO), 3.85 (br m, 1H, CHCH₂NHCO), 3.73 (s, 3H, ArOH), 3.67 (dt, 1H, *J* = 13.0, 4.5 Hz, CHCH₂NHCO), 3.60 (s, 3H, ArOH), 3.44 (s, 3H, ArOH), 3.43 (s, 3H, ArOH), 3.18–3.23 (m, 3H, ArCHCHCH₂Ar, ArCH₂CHCH₂N, ArCH₂CHCH₂N), 5 3.05 (dd, 1H, *J* = 18.0, 7.5 Hz, ArCH₂CHCH₂N), 2.90–3.00 (m, 2H, ArCHCHCH₂Ar, ArCH₂CHCH₂N), 2.71 (d, 1H, *J* = 20.0 Hz, ArCH₂CHCH₂N), 2.29 (s, 3H, NCH₃), 2.23 (dd, 1H, *J* = 15.5, 12.0 Hz, ArCHCHCH₂Ar), 2.13 (s, 3H, ArCH₃), 1.92 (s, 3H, ArCH₃); FTIR (neat film), cm⁻¹ 3364 (br, NH, OH), 1668 (s, C=O); HRMS (TOF MS ES+) *m/z* calcd for C₃₇H₄₃N₄O₇ (M+H)⁺ 655.3132, found 655.3123.

10

Example 38: Synthesis of labeled analogs:

As described generally above, certain of the inventive compounds can also be modified to permit attachment of labeling reagents. For example, certain aryl and heteroaryl groups (and other groups) as defined generically herein, can be modified (by attachment of a linker structure, 15 generally an aliphatic, heteroaliphatic, aryl, or heteroaryl moiety (or any combination thereof) which moiety is substituted or unsubstituted, cyclic or acyclic, branched or unbranched.

Methyl ester 5:



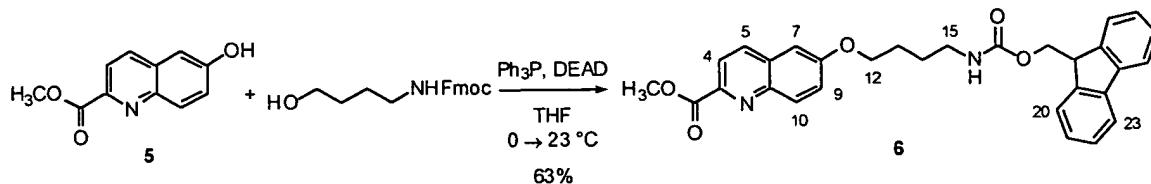
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A solution of trimethylsilyldiazomethane in hexanes (2.0M, 0.241 mL, 0.484 mmol, 2.1 equiv) was added dropwise via syringe to a suspension of acid 4 (52.0 mg, 0.231 mmol, 1 equiv) in a mixture of benzene (3.5 mL) and methanol (1.0 mL). The resulting red solution was stirred at 23 °C for 35 min. The solution was concentrated and the residual red oil was purified by flash column chromatography (25→35% ethyl acetate–hexanes). Methyl ester 5 was obtained as a white solid (44.5 mg, 94%); a bis-methylated biproduct (3.2 mg, 6%) was also isolated. Methyl ester 5: *R*_f 0.39, ethyl acetate; ¹H NMR (400 MHz, CDCl₃) δ 8.22 (d, 1H, *J* = 8.2 Hz, CHCHCOH), 8.16 (d, 1H, *J* = 8.4 Hz, CHCHCO₂CH₃), 8.13 (d, 1H, *J* = 9.2 Hz, CHCHCO₂CH₃), 7.40 (dd, 1H, *J* = 9.2, 2.8 Hz CHCHCOH), 7.17 (d, 1H, *J* = 3.2 Hz, CHCOH), 25

5.66 (br s, 1H, OH), 4.07 (s, 3H, OCH₃); ¹³C NMR (100 MHz, CD₃OD) δ 166.9, 159.3, 145.5, 143.8, 137.0, 132.8, 132.1, 124.6, 122.1, 109.1, 53.2; FTIR (neat film), cm⁻¹ 3106 (br, OH), 1726 (m, C=O), 1228 (s, Ar-OH); HMRS (CI) *m/z* calcd for C₁₁H₁₀NO₃ (M+H)⁺ 204.0661, found 204.0665.

5

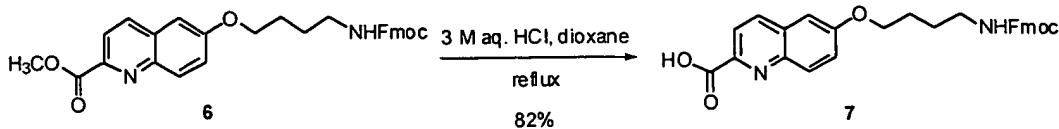
Ether 6:



Diethyl azodicarboxylate (0.255 mL, 1.62 mmol, 4.0 equiv) was added dropwise via syringe to an ice-cold solution of phenol **5** (82.2 mg, 0.405 mmol, 1 equiv), *N*-Fmoc-1-amino-4-butanol (0.504 g, 1.62 mmol, 4.0 equiv), and triphenylphosphine (0.424 g, 1.62 mmol, 4.0 equiv) in tetrahydrofuran (2.7 mL). The resulting yellow solution was allowed to warm to 23 °C and was stirred for 19 h. The solution was concentrated. Purification of the residual yellow oil by flash column chromatography (50% ether–pentane → 70% ether–pentane) afforded ether **6** as a white solid (126.8 mg, 63%): *R*_f 0.23, 50% ethyl acetate–hexanes; ¹H NMR (data given for major rotamer, 500 MHz, CDCl₃) δ 8.19 (d, 1H, *J* = 9.0 Hz, C₁₀H), 8.13–8.16 (m, 2H, C₄H, C₅H), 7.76 (d, 2H, *J* = 7.5 Hz, C₂₃H), 7.59 (d, 2H, *J* = 7.5 Hz, C₂₀H), 7.38–7.43 (m, 3H, C₂₂H, C₉H), 7.30 (t, 2H, *J* = 7.5 Hz, C₂₁H), 7.09 (d, 1H, *J* = 2.0 Hz, C₇H), 4.86 (br m, 1H, CONH), 4.43 (d, 2H, *J* = 7.0 Hz, CO₂CH₂), 4.21 (t, 1H, *J* = 6.5 Hz, CO₂CH₂CH), 4.13 (t, 2H, *J* = 5.8 Hz, C₁₂H₂), 4.07 (s, 3H, OCH₃), 3.31 (q, 2H, *J* = 6.3 Hz, C₁₅H₂), 1.91 (quint, 2H, *J* = 7.0 Hz, C₁₃H₂), 1.76 (quint, 2H, *J* = 7.5 Hz, C₁₄H₂); ¹³C NMR (100 MHz, CDCl₃) δ 166.1, 158.7, 156.5, 145.3, 143.9, 143.6, 141.3, 135.6, 132.1, 130.8, 127.6, 127.0, 124.9, 123.6, 121.4, 119.9, 105.2, 67.8, 66.5, 47.2, 40.5, 29.7, 26.7, 26.2; FTIR (neat film), cm⁻¹ 3334 (br, NH), 1720 (br s, OC=O, NHC=O), 1227 (s, Ar-OCH₂).

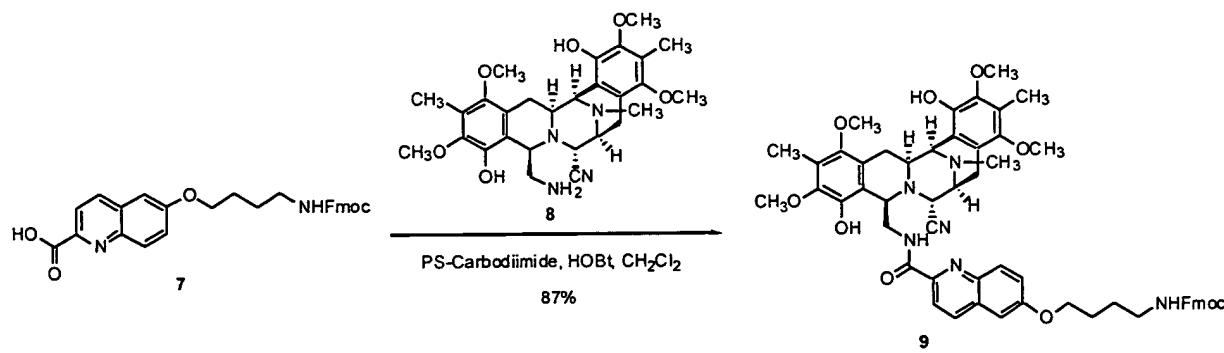
25

Carboxylic Acid 7:



A solution of methyl ester **6** (123 mg, 0.247 mmol) in a mixture of 3 M aq. HCl (0.5 mL) and *p*-dioxane (2.5 mL) was heated at reflux for 9 h. The solution was allowed to cool to 23 °C, and was concentrated. The residual yellow solid was recrystallized from ethanol:water (2:1, 30 mL). Upon cooling to –20 °C, a white amorphous solid precipitated. The solid was collected by vacuum filtration and was washed with water (2 mL), yielding acid **7** (106 mg, 82%) as a white powder: R_f 0.0, 50% ethyl acetate–hexanes; ^1H NMR (500 MHz, CD₃OD) δ 8.36 (d, 1H, J = 9.0 Hz, C₄H), 8.09–8.13 (m, 2H, C₃H, C₉H), 7.77 (d, 2H, J = 7.5 Hz, C₂₂H), 7.62 (d, 2H, J = 7.5 Hz, C₂₀H), 7.49 (dd, 1H, J = 9.5, 2.5 Hz, C₈H), 7.34–7.38 (m, 3H, C₂₁H, C₆H), 7.27 (t, 2H, J = 7.3 Hz, C₂₀H), 4.34 (d, 2H, J = 6.5 Hz, CO₂CH₂), 4.18 (t, 2H, J = 6.0 Hz, ArOCH₂), 4.16 (t, 1H, J = 6.5 Hz, CO₂CH₂CH), 3.21 (t, 2H, J = 6.5 Hz, CH₂NHCO₂), 1.88 (quint, 2H, J = 7.0 Hz, ArOCH₂CH₂), 1.72 (quint, 2H, J = 7.3 Hz, CH₂CH₂NHCO₂); ^{13}C NMR (100 MHz, CD₃OD) δ 165.7, 159.5, 157.0, 146.1, 145.2, 143.3, 142.1, 136.8, 131.9, 131.7, 128.1, 127.6, 125.5, 124.2, 121.4, 120.6, 106.4, 68.6, 48.2, 40.9, 30.3, 27.3, 26.9; FTIR (neat film), cm^{−1} 3326 (br, CO₂H, NH), 1714 (s, HOC=O, NHCO₂), 1227 (s, Ar-OCH₂); HRMS (TOF MS ES+) *m/z* calcd for C₂₉H₂₇N₂O₅ (M+H)⁺ 483.1920, found 483.1924.

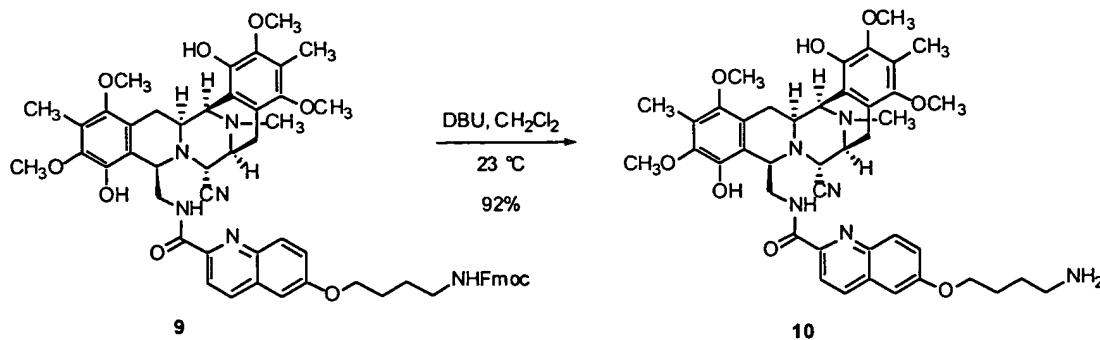
20 Amide 9:



A solution of primary amine **8** (13.8 mg, 26.3 μ mol, 1.02 equiv) in dichloromethane (0.75 mL) was added via cannula to a mixture of acid **7** (12.5 mg, 25.9 μ mol, 1 equiv) and 1-hydroxybenzotriazole (5.9 mg, 43.7 μ mol, 1.7 equiv) in an oven-dried glass vial. To the

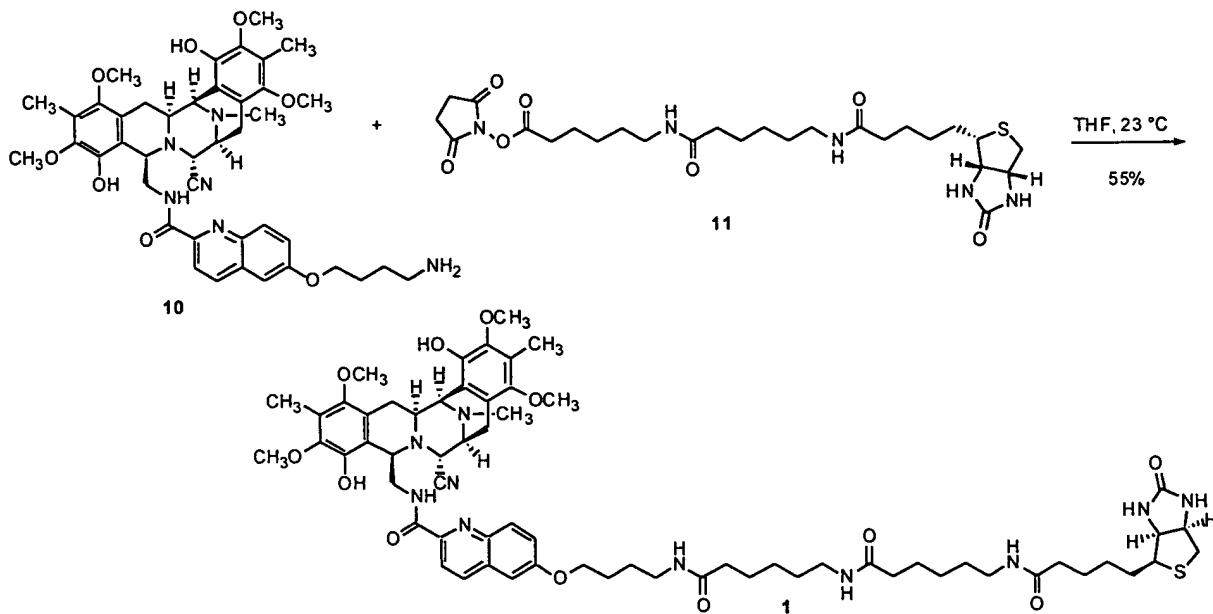
resulting suspension was added PS-carbodiimide (Argonaut Technologies, 1.39 mmol/g, 37.3 mg, 51.8 μ mol, 2.0 equiv). The resulting suspension was stirred gently at 23 °C for 19 h. The reaction mixture was directly purified by chromatography on a pipette column of silica gel (2% methanol–dichloromethane). Amide **9** was obtained as a colorless oil (22.2 mg, 87%): R_f 0.59,
 5 10% methanol–dichloromethane; ^1H NMR (500 MHz, CDCl_3) δ 8.06 (m, 2H, CHCHCCONH , CHCHCCONH), 7.90 (t, 1H, J = 6.0 Hz, CONH), 7.77 (d, 2H, J = 7.5 Hz, ArH), 7.69 (d, 1H, J = 9.5 Hz, CHCHCOCH_2), 7.60 (d, 2H, J = 7.5 Hz, ArH), 7.40 (t, 2H, J = 7.3 Hz, ArH), 7.35 (dd, 1H, J = 9.5, 2.5 Hz, CHCHCOCH_2), 7.31 (t, 2H, J = 7.5 Hz, ArH), 7.04 (d, 1H, J = 2.5 Hz, CHCOCH_2), 5.71 (s, 1H, ArOH), 5.45 (s, 1H, ArOH), 4.85 (br m, 1H, OCONH), 4.44 (d, 2H, J = 7.0 Hz, CO_2CH_2), 4.37 (t, 1H, J = 3.8 Hz, CHCH_2NHCO), 4.22 (t, 1H, J = 7.0 Hz, $\text{CO}_2\text{CH}_2\text{CH}$), 4.17–4.20 (m, 2H, $\text{CHC}\equiv\text{N}$, ArCHNCH₃), 4.14–4.18 (m, 2H, ArOCH₂), 3.75 (s, 3H, ArOCH₃), 3.69–3.75 (m, 1H, CH_2NHCO), 3.57 (s, 3H, ArOCH₃), 3.53 (s, 3H, ArOCH₃), 3.52–3.57 (m, 1H, CH_2NHCO), 3.45 (s, 3H, ArOCH₃), 3.41 (d of m, 1H, J = 7.0 Hz, $\text{CHCHC}\equiv\text{N}$), 3.32 (q, 2H, J = 6.7 Hz, CH_2NHCO_2), 3.24–3.30 (m, 2H, ArCHCHCH₂Ar,
 10 15 ArCHCHCH₂Ar), 3.07 (dd, 1H, J = 18.5, 8.0 Hz, $\text{CH}_2\text{CHCHC}\equiv\text{N}$), 2.74 (d, 1H, J = 18.5 Hz, $\text{CH}_2\text{CHCHC}\equiv\text{N}$), 2.27 (s, 3H, NCH₃), 2.19 (s, 3H, ArCH₃), 2.12 (dd, 1H, J = 16.3, 12.3 Hz, ArCHCHCH₂Ar), 1.99 (s, 3H, ArCH₃), 1.88–1.94 (m, 2H, $\text{CH}_2\text{CH}_2\text{OAr}$), 1.76–1.80 (m, 2H, $\text{CH}_2\text{CH}_2\text{NHCO}_2$); FTIR (neat film), cm^{-1} 3378 (br, NH, OH), 2233 (w, C≡N), 1714 (m, NHCO₂), 1668 (m, NHC=O); HRMS (TOF MS ES+) *m/z* calcd for $\text{C}_{57}\text{H}_{61}\text{N}_6\text{O}_{10}$ ($\text{M}+\text{H}$)⁺:
 20 989.4449, found 989.4404.

Primary Amine **10**:



1,8-Diazabicyclo[5.4.0]undec-7-ene (DBU, 0.5 μ L, 3.6 μ mol, 1.5 equiv) was added to a solution of carbamate **9** (2.4 mg, 2.4 μ mol, 1 equiv) in dichloromethane (0.1 mL). The resulting solution was stirred for 30 min. Purification of the crude reaction mixture by column chromatography on a pipette column (4% methanol–dichloromethane \rightarrow 10% methanol–dichloromethane \rightarrow 10% methanol in 98:2 dichloromethane:ammonium hydroxide), furnished primary amine **10** as a white solid (1.7 mg, 92%): R_f 0.03, 10% methanol–dichloromethane; ^1H NMR (400 MHz, CDCl_3) δ 8.06 (m, 2H, CHCHCCONH, CHCHCCONH), 7.90 (t, 1H, J = 5.2 Hz, CONH), 7.69 (d, 1H, J = 9.2 Hz, CHCHCOCH₂), 7.35 (dd, 1H, J = 9.2, 2.8 Hz, CHCHCOCH₂), 7.03 (d, 1H, J = 2.8 Hz, CHCOCH₂), 4.37 (t, 1H, J = 3.6 Hz, CHCH₂NHCO), 10 4.15–4.18 (m, 2H, CHC≡N, ArCHNCH₃), 4.11 (t, 1H, J = 6.4 Hz, ArOCH₂), 4.11 (t, 1H, J = 6.4 Hz, ArOCH₂), 3.75 (s, 3H, ArOCH₃), 3.69–3.75 (m, 1H, CH₂NHCO), 3.58 (s, 3H, ArOCH₃), 3.53–3.58 (m, 1H, CH₂NHCO), 3.53 (s, 3H, ArOCH₃), 3.46 (s, 3H, ArOCH₃), 3.40–3.43 (m, 1H, CHCHC≡N), 3.23–3.30 (m, 2H, ArCHCHCH₂Ar, ArCHCHCH₂Ar), 3.08 (dd, 1H, J = 18.4, 8.4 Hz, CH₂CHCHC≡N), 2.83 (br m, 2H, CH₂NH₂), 2.74 (d, 1H, J = 18.4 Hz, CH₂CHCHC≡N), 15 2.27 (s, 3H, NCH₃), 2.19 (s, 3H, ArCH₃), 2.12 (dd, 1H, J = 16.2, 11.8 Hz, ArCHCHCH₂Ar), 1.99 (s, 3H, ArCH₃), 1.88–1.96 (m, 2H, CH₂CH₂OAr), 1.65–1.74 (m, 2H, CH₂CH₂NH₂); FTIR (neat film), cm^{-1} 3378 (br, NH, OH), 2248 (w, C≡N), 1668 (m, NHC=O); HRMS (TOF MS ES+) m/z calcd for C₄₂H₅₁N₆O₈ (M+H)⁺: 767.3768, found: 767.3759.

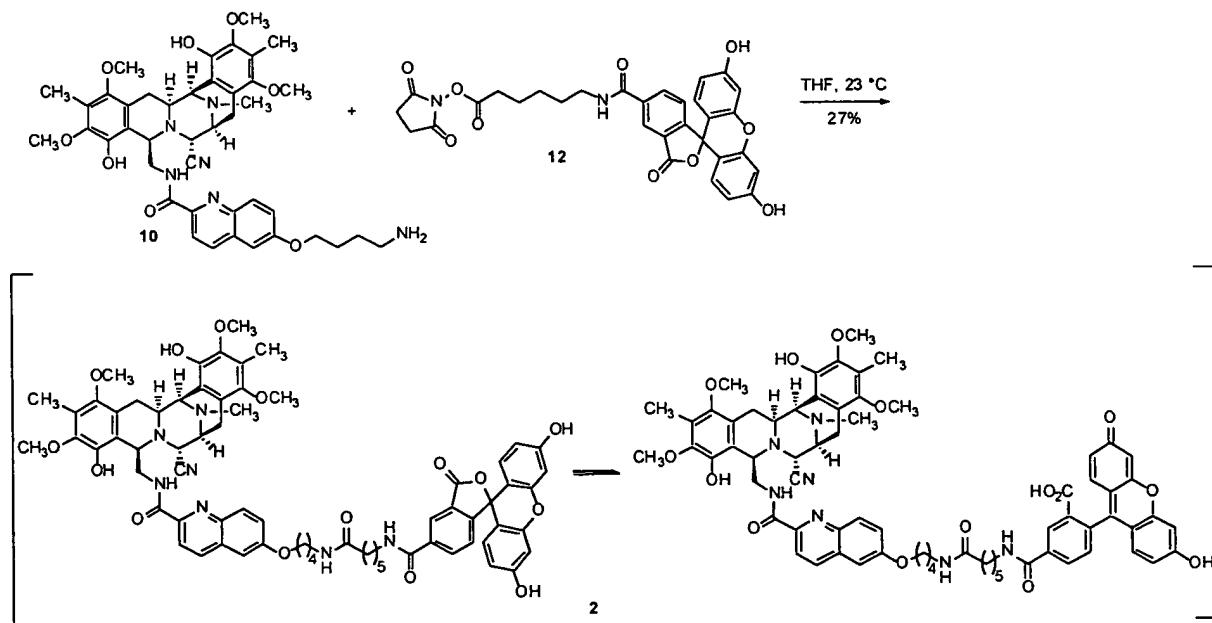
20 Biotinylated quinalascidin analog 1:



A suspension of primary amine **10** (0.8 mg, 1.0 μ mol, 1 equiv) and *N*-hydroxysuccinimidyl ester **11** (Molecular Probes, 0.71 mg, 1.3 μ mol, 1.3 equiv) in tetrahydrofuran (0.1 mL) in a glass vial was stirred at 23 °C for 14 h. The mixture was concentrated and the residual white solid was purified by column chromatography on a pipette column (5% methanol–dichloromethane \rightarrow 10% methanol–dichloromethane \rightarrow 10% methanol in 98:2 dichloromethane:ammonium hydroxide). Fractions containing the biotinylated product **1** were contaminated by remaining starting amine **10**. Further purification by chromatography on Sephadex LH-20 (11 cm \times 1.3 cm, methanol, gravity) yielded **1** as a colorless oil (0.67 mg, 55%). Material used in isolation of quinalascidin-binding proteins by affinity precipitation was further purified by HPLC (Beckman Ultrasphere ODS reverse phase column, 10 mm \times 25 mm, flow rate 2.0 mL/min, isocratic elution with 40% acetonitrile–water, retention time 29 min): R_f 0.45, 20% methanol–dichloromethane; ^1H NMR (600 MHz, CD₃OD) δ 8.18 (d, 1H, J = 8.4 Hz, CHCHCCCONH), 7.85 (d, 1H, J = 9.0 Hz, CHCHCCCONH), 7.56 (d, 1H, J = 9.0 Hz, CHCHCOCH₂), 7.40 (dd, 1H, J = 9.0, 2.4 Hz, CHCHCOCH₂), 7.23 (d, 1H, J = 2.4 Hz, CHCOCH₂), 4.44–4.48 (m, 2H, CHC≡N, SCH₂CHNH), 4.38 (br t, 1H, CHCH₂NHCO), 4.27 (dd, 1H, J = 7.8, 4.0 Hz, SCHCHNH), 4.21 (br d, 1H, ArCHNCH₃), 4.13–4.17 (m, 2H, ArOCH₂), 3.94 (dd, 1H, J = 13.8, 3.0 Hz, CHCH₂NHCO), 3.65 (s, 3H, ArOCH₃), 3.60–3.66 (m, 1H, CHCH₂NHCO), 3.43 (s, 3H, ArOCH₃), 3.41–3.46 (m, 1H, CHCHC≡N), 3.39 (s, 3H,

ArOCH₃), 3.38 (s, 3H, ArOCH₃), 3.28–3.32 (obscured by solvent peak, 1H, ArCHCHCH₂Ar), 3.27 (t, 2H, *J* = 7.2 Hz, ArOCH₂CH₂CH₂CH₂NHCO), 3.14 (t, 4H, *J* = 7.2 Hz, CH₂CH₂NHCO, CH₂CH₂NHCO), 3.13–3.18 (obscured m, SCH), 3.10 (d of m, 1H, *J* = 11.4 Hz, ArCHCHCH₂Ar), 3.03 (dd, 1H, *J* = 18.6, 8.4 Hz, CH₂CHCHC≡N), 2.89 (dd, 1H, *J* = 12.9, 5.1 Hz, SCH₂), 2.68 (d, 1H, *J* = 12.6 Hz, SCH₂), 2.61 (d, 1H, *J* = 18.6 Hz, CH₂CHCHC≡N), 2.27 (dd, 1H, *J* = 15.9, 11.7 Hz, ArCHCHCH₂Ar), 2.22 (s, 3H, NCH₃), 2.14–2.22 (m, 6H, CH₂CO, CH₂CO, CH₂CO), 2.11 (s, 3H, ArCH₃), 1.86–1.92 (m, 2H, CH₂CH₂OAr), 1.85 (s, 3H, ArCH₃), 1.70–1.76 (m, 2H, ArOCH₂CH₂CH₂), 1.26–1.70 (m, 18 H, COCH₂CH₂CH₂CH₂NH (both), CH₂CH₂CH₂CHS); LRMS (TOF MS ES+) *m/z* calcd for C₆₅H₉₁N₁₀O₁₂S (M+H)⁺ 1220, found 10 1220.

Fluorescein-labelled quinalascidin analog 2:



15

A solution of primary amine **10** (0.695 mg, 0.91 μmol, 1 equiv) and N-hydroxysuccinimidyl ester **12** (Molecular Probes, 0.560 mg, 0.96 μmol, 1.05 equiv) in tetrahydrofuran (0.2 mL) was stirred in the dark at 23 °C for 71 h. The yellow solution was concentrated. The solid residue was purified by HPLC (Beckman Ultrasphere ODS reverse phase column, 10 mm × 20 25 mm, flow rate 2.0 mL/min, gradient elution from 5% to 60% acetonitrile in water over 40

min, injected crude product as a solution in 50% acetonitrile–water (200 μ L), retention time 36 min). Fractions containing the desired product were pooled and were concentrated to remove acetonitrile. The residual aqueous solution was lyophilized, yielding fluorescein-labelled analog 2 (0.305 mg, 27%) as an orange solid: R_f 0.49, 20% methanol–dichloromethane; ^1H NMR (600 MHz, CD₃OD) δ 8.44 (br s, 1H, NHCOCHCO₂), 8.16 (d, 1H, J = 8.4 Hz, NCCHCH), 8.03 (br m, 1H, CO₂CCCHCHCONH), 7.83 (d, 1H, J = 9.0 Hz, NCCHCH), 7.54 (d, 1H, J = 8.4 Hz, CHCHCOCH₂), 7.38 (dd, 1H, J = 9.0, 2.4 Hz, CHCHCOCH₂), 7.29 (d, 1H, J = 7.8 Hz, CO₂CCCHCHCONH), 7.20 (d, 1H, J = 3.0 Hz, CHCOCH₂), 6.58–6.67 (br m, 6H, CHCHCOHCH, CHCHCOHCH), 4.45 (d, 1H, J = 2.4 Hz, CHC≡N), 4.37 (br s, 1H, CHCH₂NHCO), 4.19–4.21 (m, 1H, ArCHNCH₃), 4.11–4.14 (m, 2H, ArOCH₂), 3.94 (dd, 1H, J = 13.2, 3.0 Hz, CHCH₂NHCO), 3.64 (s, 3H, ArOCH₃), 3.48 (dd, 1H, J = 13.5, 3.3 Hz, CHCH₂NHCO), 3.44 (t, 2H, J = 7.2 Hz, CH₂NHCO), 3.42 (s, 3H, ArOCH₃), 3.38 (s, 3H, ArOCH₃), 3.37 (s, 3H, ArOCH₃), 3.28 (t, 2H, J = 7.2 Hz, CH₂NHCO), 3.09 (d of m, 1H, J = 11.4 Hz, ArCHCHCH₂Ar), 3.02 (dd, 1H, J = 18.6, 7.8 Hz, CH₂CHCHC≡N), 2.60 (d, 1H, J = 18.6 Hz, CH₂CHCHC≡N), 2.27 (dd, 1H, J = 15.6, 11.4 Hz, ArCHCHCH₂Ar), 2.24 (t, 2H, J = 7.2 Hz, CH₂CONH), 2.22 (s, 3H, NCH₃), 2.10 (s, 3H, ArCH₃), 1.85–1.90 (m, 2H, CH₂CH₂OAr), 1.83 (s, 3H, ArCH₃), 1.66–1.76 (m, 4H, CH₂CH₂NHCO, CH₂CH₂NHCO); LRMS (TOF MS ES+) *m/z* calcd for C₆₉H₇₂N₇O₁₅ (M+H)⁺ 1239, found 1239.

20 **IV. In vitro activity:**

a) Experimentals:

Cells and Cell Culture Conditions

25 The two cell lines used, A375 malignant melanoma and A-549 lung carcinoma, were purchased from American Type Culture Collection. These cells were cultured at 37 °C in a humidified atmosphere of 5% CO₂ in Dulbecco's Modified Eagle Medium (DMEM) supplemented with 10% fetal bovine serum (FBS), 0.1% glutamine and 0.1% penicillin-streptomycin. DMEM, FBS, glutamine and penicillin-streptomycin were purchased from Life
30 Technologies (Grand Island, NY).

Cell Growth Inhibition Assay

Exponentially growing cells were seeded at ~3000 (50 μ l) cells per well in 96-well flat-bottomed microtiter plates and then incubated at 37 °C in a humidified atmosphere of 5% CO₂ / 5 95% air for 24 hr. An analogue was dissolved in dimethyl sulfoxide to give a concentration of 0.6 mg/ml, which was further diluted with the culture medium containing 10% fetal bovine serum. Nine three-fold dilutions were prepared with the maximum concentration being 1/300 of the original DMSO solution (i.e. 2000 ng/ml). This procedure was repeated for each analogue. Fifty microliters of the obtained dilutions were each transferred into the well of the above 10 described culture plate. The cell culture was then placed back into the incubator at 37°C under the 5 % CO₂ atmosphere for 72 hr. Cell proliferation was quantified by using the CellTiter 96 AQueous Assay (Promega). In this assay a mixture solution (20 μ l) of MTS [Owen's reagent: 3-(4,5-dimethylthiazol-2-yl)-5-(3-carbomethoxyphenyl)-2-(4-sulfophenyl)-2H-tetrazolium inner salt] and phenazine methosulfate (PMS) was added to each well. The resulting mixtures were 15 further incubated for 2.5 hr. The absorbance was measured with a microplate reader at a test wavelength of 540 nm and a reference wavelength of 655 nm to serve as an index of the number of viable cells. The inhibitory ratio of the test compound was calculated according to the following formula: inhibition ratio (%) = 100 x (C – T) / C, where T is an absorbance of the well containing a test compound and C is an absorbance of the well containing no test compound. The 20 IC₅₀ was calculated by the least squares method.

b) **Exemplary Data:**

Table 1 ($N = R_1$ in generic structure, where m is 1)

Table 1 (continued)

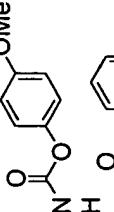
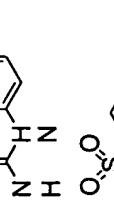
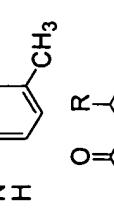
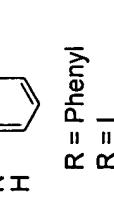
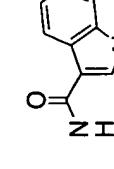
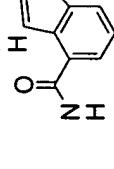
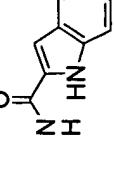
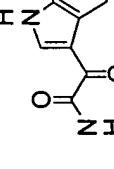
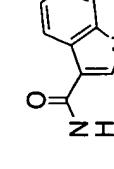
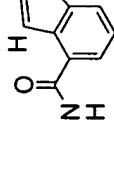
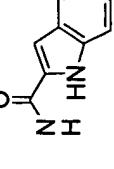
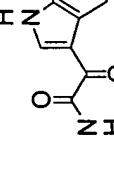
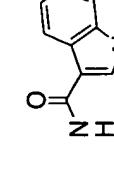
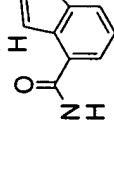
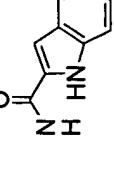
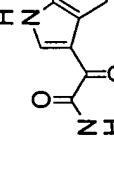
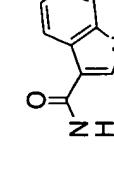
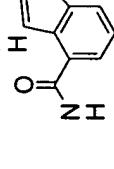
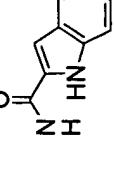
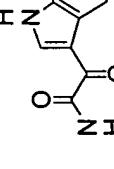
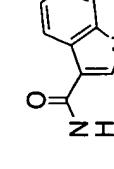
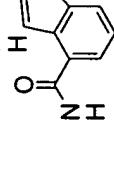
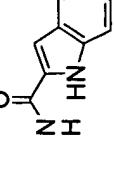
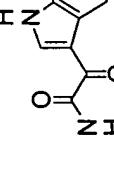
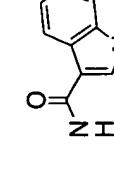
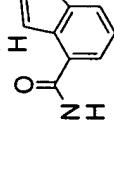
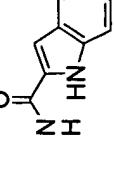
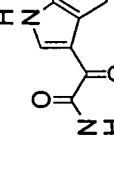
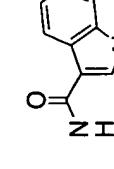
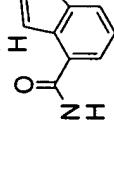
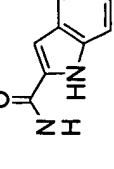
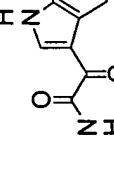
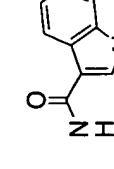
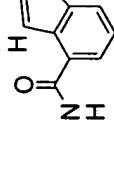
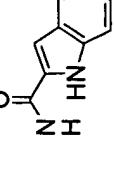
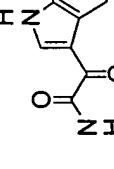
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	A375	A549	A375	A549		A375	A549		A375	A549	
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	37	340		85		81	1100		R = CH ₃	2.5	4.0
	81	860		860		2.5	26		R = OCH ₃	9.6	3.5
	2.5	37		9.6		96	1100		R = Cl	11	4.1
	2.5	14		96		96	>1400		(15)	1.2	4.7
	2.5	1.4		310		310	>510		(16) ¹⁰	3.6	78
	8	1.2		11.3		11.3	1.7		(10)	9.2	8.3
	8	1.2		6.5		6.5	1.7		(16) ¹⁰	8.3	200
	9	1.2		4.1		4.1	99		(diastereoisomers)	42	1100
										34	975

Table 2

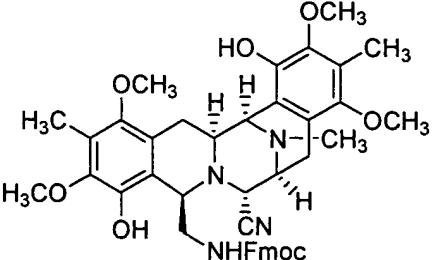
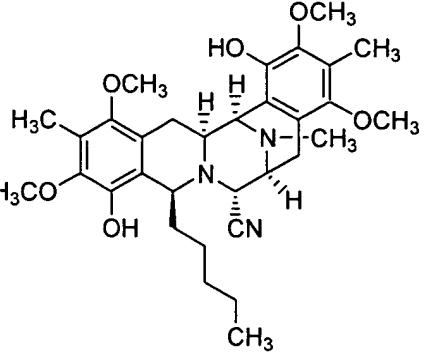
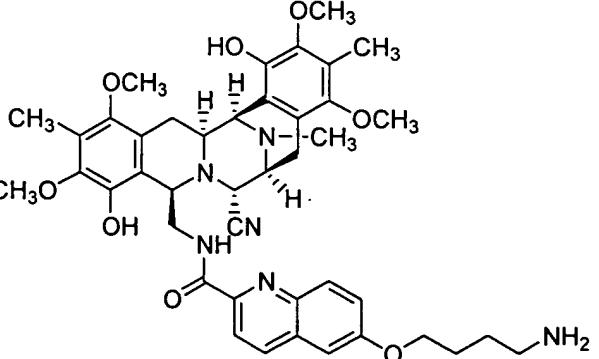
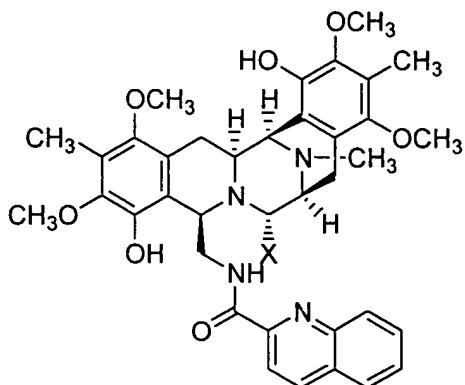
	IC ₅₀ (nM)	Cell line
A375-Melanoma		A549-Lung
	35	176
	116	1400
	29	118
Biotinylated derivative	210	990
Fluorescein derivative	18	590

Table 3



X	IC ₅₀ (nM)	
	Cell line	
OH	0.9	13
OCH ₃	120	1100
H	>9000	>9000

V. In vivo activity:

Although a variety of methods can be utilized, one exemplary method by which the *in vivo* activity of the inventive compounds is determined is by subcutaneously transplanting a
5 desired tumor mass in mice. Drug treatment is then initiated when tumor mass reaches approximately 100 mm³ after transplantation of the tumor mass. A suitable composition, as described in more detail above, is then administered to the mice, preferably in saline and also preferably administered once a day at doses of 5, 10 and 25 mg/kg, although it will be appreciated that other doses can also be administered. Body weight and tumor size are then
10 measured daily and changes in percent ratio to initial values are plotted. In cases where the transplanted tumor ulcerates, the weight loss exceeds 25-30% of control weight loss, the tumor weight reaches 10% of the body weight of the cancer-bearing mouse, or the cancer-bearing mouse is dying, the animal is sacrificed in accordance with guidelines for animal welfare.